POWELL

VOLUME XXII DECEMBER, 1939 NUMBER 12

JOURNAL OF DAIRY SCIENCE

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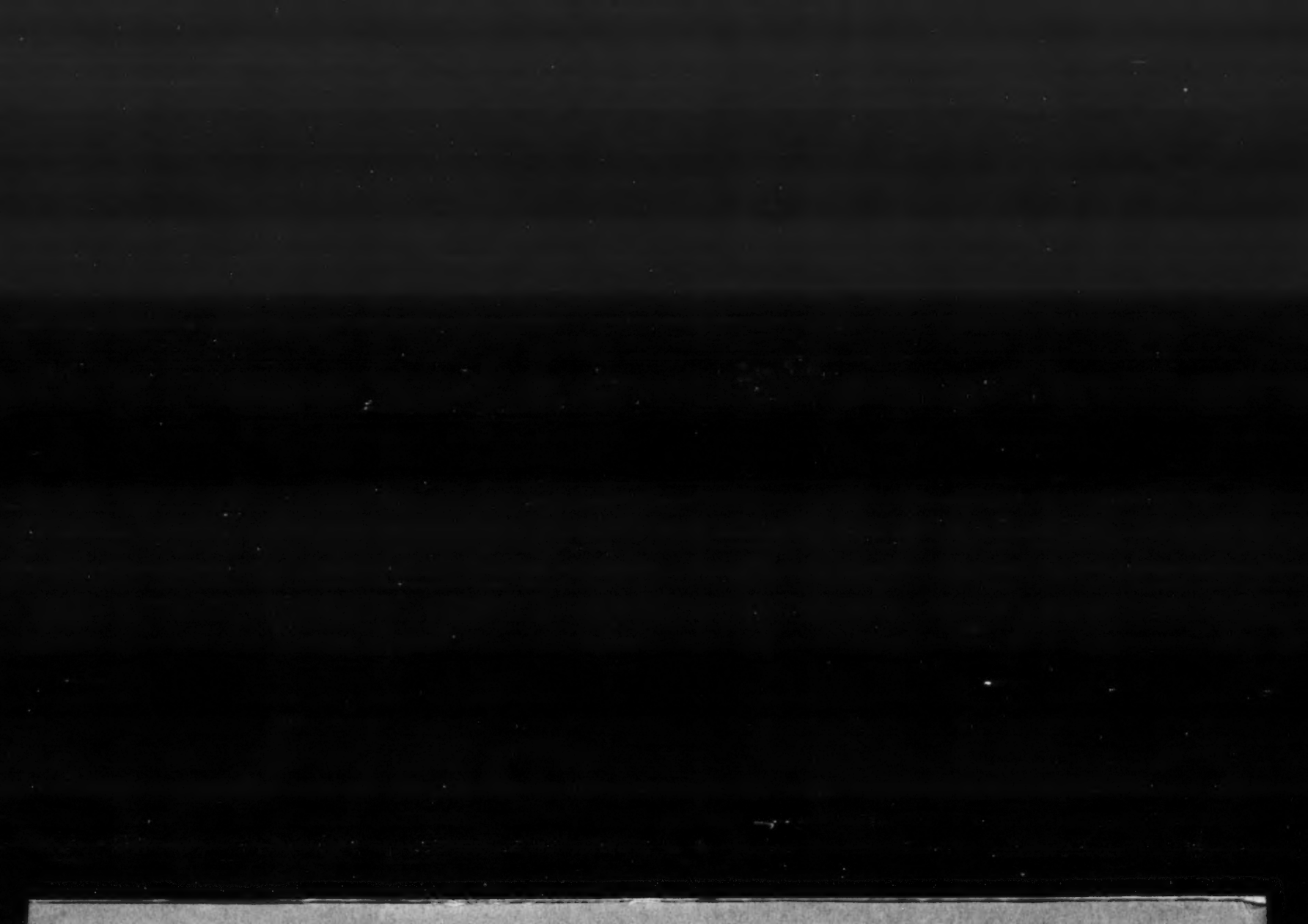

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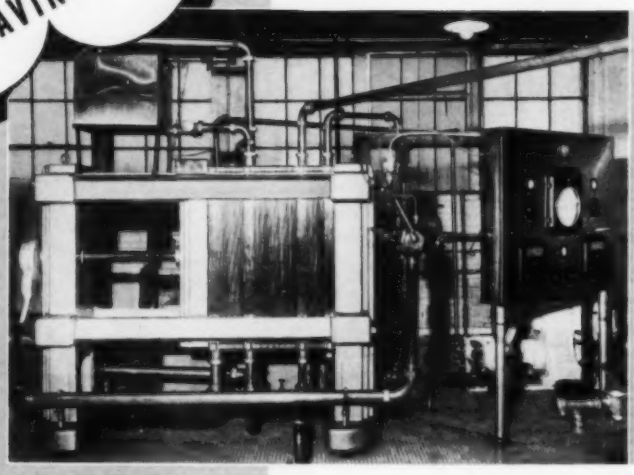

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JOURNAL OF DAIRY SCIENCE

VOLUME XXII DECEMBER, 1939 NUMBER 12

A STUDY OF THE pH VALUES OF THE INGESTA OF THE BOVINE RUMEN

C. F. MONROE AND A. E. PERKINS[1]

Department of Dairy Industry, Ohio Agricultural Experiment Station, Wooster, Ohio

The rumen, or first compartment of the ruminant stomach, plays a very important role in the digestive processes of the bovine species. It is here that the feed after being swallowed is stored, mixed, and softened with water and saliva, as well as subjected to a certain amount of digestive activity by bacteria. To maintain the optimum conditions for the bacterial activity, it is logical to assume that one of the requirements would be the maintenance of a proper hydrogen ion concentration. Although nature probably has a means of regulating the reaction in the rumen, it is probable that extremes in feeding have some influence. For instance, such extremes as heavy grain feeding or the excessive use of corn or A.I.V. silages may affect the balance.

LITERATURE

Dukes (1) states that "the reaction of the rumen content of the ox is always alkaline; the average is pH 8.89 (Gabriel)." He explains this constantly alkaline reaction on the basis of the addition of the highly alkaline saliva secreted by ruminants. An alkaline rumen reaction is noted by Stalfors (3) when the roughage fed was hay, straw, and oats bran or hay alone. Schwarz and Stremnitzer (4) found an average pH value of 8.28 in eleven animals that had been slaughtered. The day previous to slaughter these animals were fed exclusively on hay and straw. Kick and associates (2) working with steers having rumen fistulas report that "the pH of the ingesta varied from 5.5 to 7.7 depending on the ration." The most alkaline conditions were encountered when alfalfa hay alone was fed. They also report that there were variations in reactions during the day.

Received for publication May 11, 1939.

[1] The authors wish to express appreciation for the advice and assistance given by Dr. A. F. Schalk of the Veterinary College of the Ohio State University, who is also affiliated with the Animal Industry Department of this Station. Likewise, credit is due to Dr. C. H. Kick for timely suggestions at the beginning of this work. Dr. Kick now deceased, was formerly, associated with the Animal Industry Department of this Station, and later was head of the Department of Animal Husbandry of Colorado State College. We are indebted to Mr. Stanley Powers of our Station for drawing the graphs.

METHOD

The samples of ingesta used in this study were obtained from animals in which a permanent rumen fistula had been established by surgical operation. Two such animals were used, one a Holstein steer approximately 18 months of age and the other a Jersey cow in her first lactation period. With the exception of having the fistula these animals were apparently normal. No special difficulty was encountered in maintaining the animals in a fairly thrifty condition. The steer gained in live weight as well as his feeding would permit, and the cow, although never a heavy milker, produced fairly well. She calved and conceived again during the course of the work. Both animals were kept in box stalls and bedded with wood shavings. When the experimental procedure called for it, they were turned out to pasture as normal animals. Neither animal gave any signs of undue discomfort when the samples were taken. The fistulas were kept closed by means of rubber plugs at all times except for the brief periods when the samples were being taken.

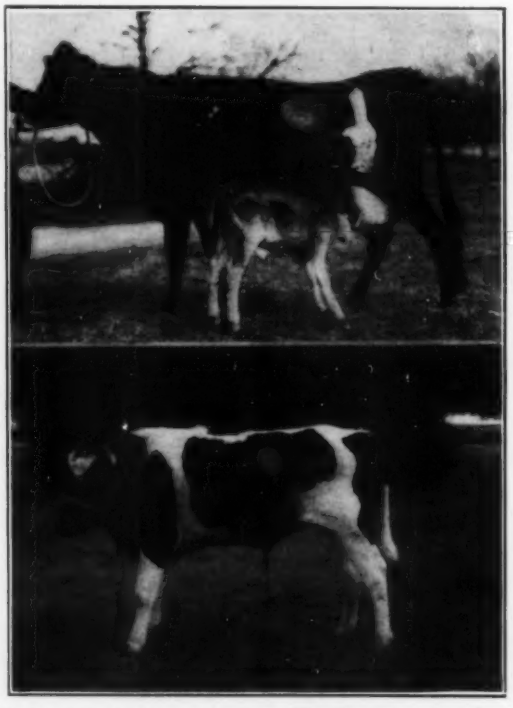

FIG. 1. Animals used in the experiments. The Jersey cow was bred and freshened during the investigation. The fistulas were kept closed by special rubber plugs which consisted of inner and outer flanges joined in the center by a core approximating the size of the opening.

In sampling the contents of the rumen six samples were taken each time. These were obtained from different locations as follows: three from the upper, or dorsal portion, distributed from front to rear, and three from the bottom, or ventral portion, from front to rear. Hereafter the top samples will be designated as front, middle, and rear, whereas the lower set will be designated as deep front, deep middle, and deep rear. The upper samples were taken approximately 1 or 2 inches below the surface of the rumen contents and the deep samples were taken at about the same distance from the floor of the rumen. In reporting the pH values, the average of the six readings is given unless otherwise specified.

When the samples were taken only once during the day, this was done at 1 P.M. Later, additional samplings were made at 2-hour intervals starting at 5 A.M. and finishing at 5 P.M. It was believed that this frequent sampling would tell a more complete story than the one-time-a-day sampling, although this latter method would have merit for comparative purposes.

Fortunately, as shown by the 2-hour procedure, the choice of 1 P.M. as a sampling hour was good, because at this time the pH reaction was close to the point of equilibrium for the day.

The pH values were determined on the uncentrifuged liquid expressed from the ingesta. All determinations were made on the fresh material within 1 hour or less after the samples were taken. The readings were made on a Leeds and Northrup portable potentiometer with a saturated calomel half cell and a quinhydrone electrode.

RESULTS

The pH values of the one-time-a-day samplings are shown in table 1. This table also shows the dates of sampling and the rations, together with the feeding periods. The average pH values of the urine are given in those cases where a sufficient number of urine samples was taken to justify an average. The data are classified according to the rations fed rather than in chronological order. It is believed that the animals were fed the different rations over periods sufficiently long for an equilibrium to be established.

When hay and grain were fed, the average pH value of the ingesta was 7.01. These results obtained in three different periods were fairly consistent. In the one period when both animals received this same ration the averages were very close together. The urine was consistently alkaline, averaging pH 8.29 for all the periods. In this feeding the amounts of hay given were either double or a little more than double the amounts of grain.

The addition of corn silage to the hay-grain ration and a slight reduction in hay did not result in a significant change in the pH values of the ingesta. The pH values on the hay-grain-corn silage ration averaged 6.95. Unfortunately, urine samples were not consistently taken on this ration, but those which were obtained at irregular intervals averaged a little over 8.00.

When A.I.V. silage having a pH value of 3.66 was substituted for the corn silage in the ration of the steer the pH value of the ingesta was little if any changed. The average was in this case 6.98. The urine pH value was lowered to around 6.35, however.

TABLE 1

The pH values of ingesta from various rations. Data from one-time-a-day samplings (Average pH values for urine shown in some cases)

Daily ration (pounds)				Experimental period	Experimental animal	Number of trials	pH values		
		Silage					Ingesta		Urine
Grain*	Hay†	Corn	A.I.V.‡				Range	Av.	Av.
				Days					
5	10	.....	.....	26	Steer	5	6.90–7.08	6.99	8.41
6	12	.....	.....	26	Cow	5	6.85–7.13	7.02	8.32
6	14	.....	.....	106	Cow	5	6.88–7.40	7.14	.....
8	18	.....	.....	84	Cow	9	6.54–7.04	6.89	8.13
					Average for ration		.....	7.01	8.29
4	4	12	.....	39	Steer	10	6.74–7.16	6.94	.....
4	4	12	.....	29	Steer	5	6.81–7.03	6.91	.....
6	6	12	.....	29	Cow	5	6.75–7.05	6.93	.....
10	14	20	.....	32	Cow	4	6.91–7.20	7.03	.....
					Average for ration		.....	6.95	.....
4	4	.....	12	34	Steer	7	6.90–7.09	6.98	6.35
4	.....	12	12	31	Steer	7	6.60–7.00	6.87	5.91
4	..... (Plus 2 oz. limestone)	12	12	13	Steer	3	6.78–7.07	6.97	5.73
10	.....	68	.....	47	Cow	10	6.58–7.10	6.83	6.59
10	Straw 2	56	.....	14	Cow	5	6.76–7.04	6.90	6.36
5	Bluegrass pasture		.....	28	Steer	8	5.91–6.86	6.44	.....
6	" "		.....	28	Cow	8	6.06–6.92	6.50	.....
					Average for ration		.....	6.47	.....
6	Alfalfa pasture		.....	48	Cow	5	6.55–6.85	6.66	.....

* Grain mixture { Corn 500 lb. | Oats 300 lb. | Wheat bran 100 lb. | Linseed Oilmeal 100 lb. | Salt 9 lb.

† Hay, Second-cutting alfalfa.

‡ A.I.V. silage was made out of first-cutting alfalfa, containing some clover and timothy.

A still further change in the ration involving the substitution of corn silage for the hay made but little change in the reaction of the ingesta. On this ration, consisting of grain, corn silage, and A.I.V. silage, the reaction was 6.87. In the following period when 2 ounces of ground limestone were fed daily, the pH value was 6.97. The reaction of the urine on this ration with and without the supplemental limestone feeding was acid.

A ration of grain with corn silage as the only roughage gave a pH reaction

of 6.83. The daily addition of 2 pounds of straw had but little effect. The pH in this case was 6.90. The urine was also acid on this ration.

On both bluegrass and alfalfa pasture the pH values of the ingesta were more acid than on any of the other rations.

An average pH value of 6.47 was obtained on bluegrass pasture and 6.66 on the alfalfa pasture. Although urine samples were not consistently taken, those that were taken when the animals were on the pastures show an average of around pH 8.00.

Two-hour sampling

All the results obtained in this phase of the work were obtained from the cow. The data are shown by means of graphs, in the accompanying chart. In addition, there is shown in table 2 a complete set of readings for one of the rations.

The table represents averages for three repetitions taken a week or more apart and after the animal had been on the ration a sufficient length of time to become adjusted to it. This is true for the other rations shown in the graphs with the following exceptions. The graph for the grain-corn silage-straw ration is for one day's collection and that for the grain and pasture is for two days' collection.

These curves, with the exception of the one on the pasture ration, show the same general trend. Before feeding in the morning, at 5 A.M., the reaction was at the most alkaline point of the day. After feeding there was a change to an acid condition which reached the low point at 9 A.M. There was then a gradual upward trend until 3 P.M., or just before the afternoon feeding. After this feeding there was another drop in the reaction.

The curve for the pasture-grain ration is much different from those for the other rations. The reactions show less variability during the day. From 5 A.M. to 11 A.M. there was little change. At 1 P.M. there was a slight drop in reaction, and then from 1 to 5 P.M. the readings were very close together.

The physical condition of the contents of the rumen was much softer and more liquid when the cow was on pasture than when she received hay and the silage combinations in the barn.

The values obtained by sampling every 2 hours during the day agree fairly well with those obtained by sampling once a day at 1 P.M. The agreement falls within 0.25 pH.

The trend of the reactions shown by the curve from an acid condition following eating toward an alkaline condition is probably the result of two factors. First, there is the copious secretion of alkaline saliva passing from the mouth into the rumen. Dukes (1) estimates that the ox secretes daily 56,000 grams of saliva. The same authority quotes figures of 8.1 and 8.23 as the pH value of this fluid. Samples of saliva taken from the experimental animals used in this work have shown a pH of 8.0 to 8.4. Second, there is the elimination of at least part of the acids, probably by their being dissolved out

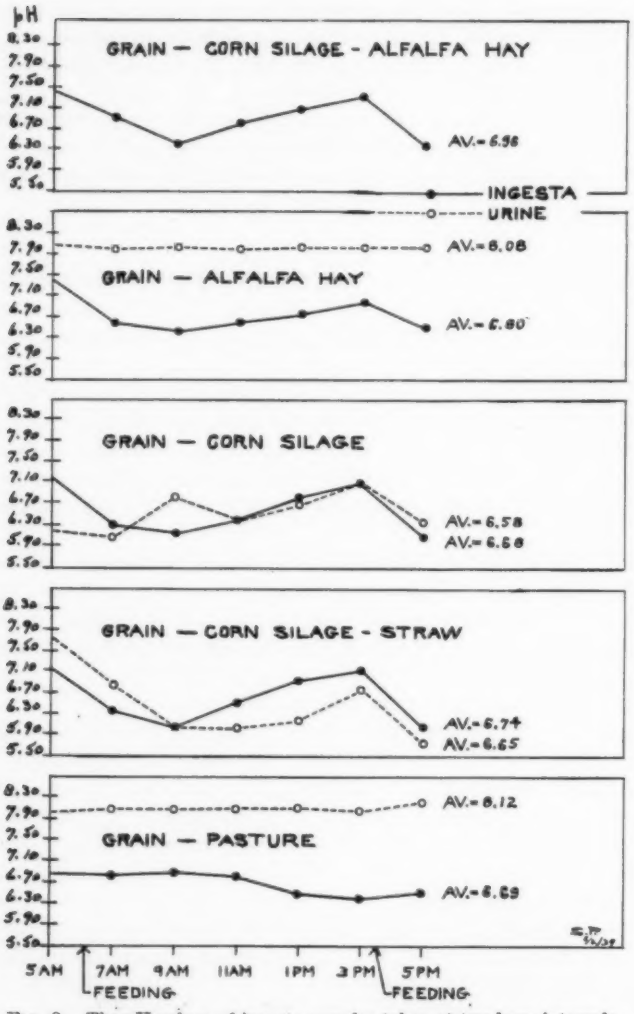

FIG. 2. The pH values of ingesta samples taken at two-hour intervals.

and passed along the alimentary canal with the more fluid portion of the ingesta. The change in urine reactions during the course of the day when acid rations were fed would indicate that this process goes on.

That the changes in reaction are the result of activity within the animal would be indicated by the following observations. Samples of ingesta taken at the 7 A.M. collection were kept in covered Mason jars in an incubator at

TABLE 2

pH values of ingesta, taken at 2-hour intervals. (Daily ration fed cow: 10 pounds of grain, 60 to 68 pounds of corn silage) (composite of three sampling dates, 2/23, 3/5 and 3/18)

Sample	Time							
	5 A.M.	7 A.M.	9 A.M.	11 A.M.	1 P.M.	3 P.M.	5 P.M.	Av.
	pH	*pH*	*pH*	*pH*	*pH*	*pH*	*pH*	*pH*
Front	7.13	6.00	5.79	6.01	6.76	7.08	5.89	6.46
Middle	7.21	6.15	6.04	6.07	6.87	7.12	6.01	6.58
Rear	7.24	6.21	6.19	6.51	6.83	7.22	6.18	6.70
Deep front	7.17	6.51	6.37	6.80	6.90	7.06	6.23	6.80
Deep middle	7.21	6.48	6.40	6.60	6.91	7.09	6.37	6.78
Deep rear	7.19	6.42	6.39	6.42	6.86	7.03	6.33	6.72
Av.	7.19	6.30	6.20	6.40	6.86	7.10	6.17	.....
Urine	6.27	6.18	6.82	6.38	6.70	7.10	6.32	.....

Av. for Day—Ingesta, pH 6.68; Urine, pH 6.58.

100° F. for approximately 8 hours. A comparison of the pH values of this material after 8 hours with those of the original ingesta and of the material which had been allowed to remain in the rumen for this time showed that the acidity of the material in the jars increased rather than decreased. (See table 3.)

TABLE 3

Comparison of pH values taken at 7 A.M. and incubated for 8 hours with the same material allowed to remain in the cow

Sample*	pH values		
	At 7 A.M. (from cow)	At 3 P.M. (from cow)	Taken at 7 A.M. and incubated at 100° F. until 3 P.M.
1	6.92	7.24	5.17
2	6.83	7.29	5.36
3	6.98	7.13	5.47
4	6.30	7.31	5.71
5	6.39	7.02	5.51
6	6.09	6.39	5.54
7	6.37	7.12	6.40

* Samples taken on different days, various rations fed. Other samples taken at 9 A.M. and 11 A.M. showed the same trend.

pH reactions of different parts of the paunch

From the data presented in table 2, it will be noted that the reaction of the ingesta is not uniform throughout the rumen. This is probably a matter of mixing. The longer the material had been in the paunch the better it was mixed and the more nearly uniform were the reactions. For such a study the average reactions of the number of different readings have been compared.

The data in table 4 indicate that the reactions for the lower part of the paunch were fairly uniform and more alkaline than those of the upper por-

TABLE 4

Reactions of ingesta in different parts of the paunch at 5 A.M. and 9 A.M. (Data taken from 2-hour interval samples, pasture periods not included)

Time	Front	Middle	Rear	Deep front	Deep middle	Deep rear	Av.
5 A.M.	7.23	7.36	7.32	7.28	7.32	7.31	7.31
9 A.M.	6.00	6.07	6.24	6.47	6.44	6.45	6.28
Av. for day	6.58	6.70	6.81	6.90	6.90	6.89	6.80
From one-time-a-day samples. Taken at 1 P.M. (Same type of rations as above, but not same samples)							
	6.85	6.94	7.02	6.98	6.99	6.99	6.96

tion. Of the six different locations, the front was the most acid. The variation in reactions ranged from 0.13 pH at 5 A.M. to 0.47 pH at 9 A.M. The range in the values for the average of the day was 0.32, and for the one-time-a-day samples taken at 1 P.M., 0.14 pH.

Slaughter data

An opportunity for obtaining the reactions on slaughtered animals occurred when it became necessary to kill a group of heifers and cows including cow 497—the Jersey cow used in this work. The animals were killed one at a time in a near-by slaughterhouse where it was possible by the aid of a portable potentiometer to make the pH determinations within 10 minutes following death. These animals had received no feed for a period of 12 to 18 hours previous to slaughter. The results of this study are shown in table 5.

TABLE 5

The pH values of the rumen contents of slaughtered animals

Animal no.	Age yrs.	Breed	Ration, pounds per day	pH values
346	11.0	Holstein	Grain 8; corn sil. 24; alf. h. 16	7.37
423	7.0	Jersey	" 8; " " 36; " " 8 and pasture.	7.30
497	4.0	"	" 4; corn sil. 20; alf. h. 10	7.42
509	3.5	Holstein	" 8; " " 20; " " 20	7.34
531	2.5	Jersey	pasture	7.34
534	2.5	"	Grain 6; corn sil. 14; alf. h. 10	7.25
537	2.5	"	" " 10 and pasture.	7.13
538	2.5	"	Grain 8; corn sil. 16; alf. h. 14	7.61
542	2.0	"	Alfalfa h. ad lib. pasture	7.30
544	2.0	"	" " " " "	7.50
545	2.0	Holstein	" " " " "	7.34
547	2.0	"	" " " " "	7.20
563	1.5	Jersey	" " " " "	7.42
564	1.5	Holstein	" " " " "	7.42
571	1.0	"	" " " " "	7.22
575	1.0	"	" " " " "	7.25
Average			Average of all,	7.34
Older animals on grain, corn silage, and hay ration			7.35	
Young animals alfalfa hay and late fall pasture			7.33	

The values obtained in the slaughter method are higher than the averages obtained by the fistula method, but they are only slightly higher than the values obtained at 5 A.M. in the 2-hour samplings. The slaughter data represent the same partially empty rumen condition as that found at 5 A.M. previous to the morning feeding. In fact an even longer interval had elapsed after feeding and before the cows were slaughtered than in the normal routine as represented in the 2-hour data. Just how much the excitement of moving and the strange quarters affected the reactions is impossible to state. Under these conditions animals are not as contented as normally and ordinarily ruminate but very little. Some appear to be more affected by the change than others.

SUMMARY

A study was made of the pH values of the contents of the bovine rumen. Samples of ingesta were obtained from animals having a rumen fistula.

The pH values of the ingesta averaged between 6.83 and 7.01 when such roughages as corn or A.I.V. silage and alfalfa hay in fair amounts were fed. On pasture, both bluegrass and alfalfa, the ingesta were a little more acid.

Samples taken at 2-hour intervals throughout the day showed the ingesta most alkaline just prior to feeding in the morning. After feeding there was a drop until about 9 A.M., and then a gradual trend toward alkalinity reaching a peak at 3 P.M. immediately preceding the evening feeding. After feeding there was again a drop in pH. This was for cows on winter rations. Those on pasture did not show these fluctuations.

The pH values were not the same in all parts of the rumen. The largest variations were found soon after feeding. At 1 P.M. these variations fell within 0.2 pH.

Samples of the rumen contents of animals slaughtered 12 to 18 hours after the last feeding had an average pH value of 7.34. These values were uniformly more alkaline than the averages noted above as taken from the living animals. They were, however, quite similar to those obtained at the 5 A.M. sampling in the 2-hour collection study.

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INJURY OR INFECTION IN UDDERS OF HEIFERS MAY TERMINATE IN BLIND QUARTERS

W. W. SWETT
Senior Dairy Husbandman
C. A. MATTHEWS
Assistant Dairy Husbandman
AND R. R. GRAVES
Chief, Division Dairy Cattle Breeding, Feeding and Management Investigation, Bureau of Dairy Industry

Marked reduction in, or complete cessation of milk secretion in one or more quarters of the udder, is not uncommon in cows. In some cases certain quarters are "blind" throughout the entire producing life of a cow. In some States and in some cities health authorities do not permit the sale of milk from cows having an inactive quarter regardless of the cause, and in some areas such cows are not permitted to remain in the herd for breeding purposes. A better understanding of the causes and the significance of such abnormalities is much to be desired.

It has been shown (1) that the four quarters of the udder develop from entirely independent tubular formations which are present and can be detected by palpating the udder of the calf at or soon after birth. These tubular formations which extend upward from the teats toward the abdominal wall first enlarge by bulging near the center. Later each of these elongated enlargements develops into a somewhat rounded formation which has no defi-

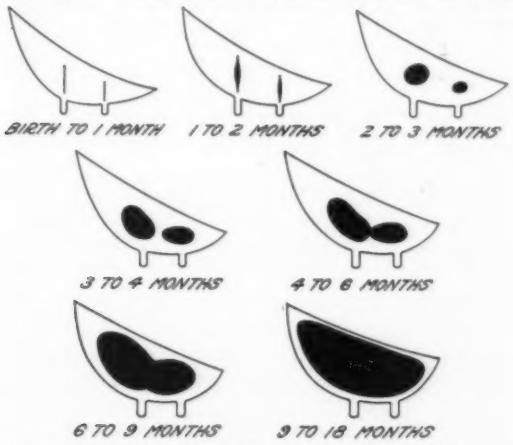

FIG. 1. Diagrammatic illustration of the manner in which the mammary gland tissue develops in a calf.

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nite connection with the corresponding development in any other quarter until at usually about 4 or 5 months of age the front and rear quarters on the same side approach and come in contact with each other at the base and subsequently appear to be entirely fused together. The secretory and ductal systems, however, remain independent of each other through life. Figure 1 shows diagramatically what may be expected in the way of glandular development in the udder of a heifer of one of the dairy breeds at the ages given.

It has long been known, although perhaps not generally realized by laymen, that the four quarters of a cow's udder are entirely separate from each other and that normally the secretory system of one does not communicate with that of another—even the one on the same side. There is a distinct septum separating the right and left halves of the udder from each other. There is no visible septum separating the front quarter from the rear one although it has been demonstrated that fluid injected into the teat canal does not pass into any of the other quarters. Figure 2 shows a horizontal trans-

FIG. 2. Horizontal transverse section through an udder in which the right front and left rear quarters had been filled with a staining solution. Each of the quarters was separate from the others.

verse section through the udder of cow No. 50. After the udder was amputated, a staining solution was injected through the teats into the right front and left rear quarters. The other two quarters were filled with uncolored formalin. The section cut from the frozen udder shows clearly that the four quarters of the udder are entirely separate from each other.

Considering the independent development of the different quarters it is not difficult to visualize how one quarter can be injured or destroyed without the others becoming affected. Figure 3 shows a section through the two front quarters of an udder, the secreting activity of the left front quarter of which

FIG. 3. Vertical transverse section through the front quarters of an udder showing that one quarter may become permanently "blind" without the other quarters being affected.

had been destroyed. The other three quarters apparently were functioning nomally.

The destruction of one quarter of an udder apparently does not necessarily impair the activity of the other quarters. In fact definite evidence has been presented elsewhere (2) to indicate that in certain cases where the loss of one or more quarters occurs early in life, there is a compensatory effect, which results in the remaining quarters becoming more than normally active in the secretion of milk.

Undoubtedly there are many different causes of "blind" or impaired quarters. Self-sucking or sucking by other calves, bruises and other injuries to the udder or teats, and mastitis, are perhaps among the most frequent causes. Many cases occur for which the cause cannot readily be determined. In the breeding herd of the Bureau of Dairy Industry at Beltsville, Md., it is the practice to make frequent examinations of the mammary gland development in calves, and of the udder structure through life in cows. Among the hundreds of udders studied over a period of years, there have been a number of cases of infection in one or more quarters that were contracted early in life. In nearly every such case the functioning of the affected quarters was permanently impaired or destroyed. Most of them failed entirely to secrete milk. Following are discussions of the history and performance of some of the observed cases, together with such photographic records as are available.

The youngest udder infection recorded is that of No. 1089. At the age of 18 days the left rear quarter was reported as being greatly enlarged and discharging pus from the teat. At 29 days the condition was essentially the same. At 1 month 28 days it was enlarged externally but the glandular development was somewhat retarded. The retarded glandular development in this quarter was marked until 4 months of age, after which the development became fairly uniform, and at 12 months was well above the breed average. When she calved no milk could be obtained from the left rear quarter. At an examination 3 days after calving the udder was swollen and no abnormality could be detected. Three weeks later, however, the left rear quarter was found to be light, stringy and apparently blind. As a 4-year-old she produced 8,808 pounds of milk and 537 pounds of butterfat from the three remaining quarters. This cow is still in the herd.

Another udder infection that occurred at a very early age is that of No. 1245. At 1 month of age the right front quarter was definitely enlarged and discharging pus. This calf was in an individual pen and could not have been sucked by other calves. Inquiry revealed the fact that the calf had jumped over the high concrete wall of her pen a few days before the condition noted above was seen. In doing so she may have bruised her udder. At 2, 3 and 4 months this quarter was definitely abnormal in development. At first it appeared enlarged and later was described as retarded. At 5 months and at

subsequent examinations the udder development appeared to be fairly uniform in the different quarters and compared favorably with the breed average for development at most ages. After calving no milk could be obtained from either the right front or the left rear quarters. At the first observation after calving both of these quarters were found to be abnormal and it was soon apparent that they were "blind." There is nothing in the records to explain the reason for the failure of the left rear quarter to function. She was milked for about 3 months and then slaughtered. The best production for one month was 922 pounds of milk and 31.9 pounds of butterfat. Figure 4A shows the appearance of the gross anatomy of the two front quarters and 4B shows the structure of the two rear quarters at the time of slaughter. In the left rear quarter much of the tissue appears to be definitely fibrous and incapable of milk secretion. The small area of fairly normal appearance near the center of this quarter apparently was a part of the left front quarter

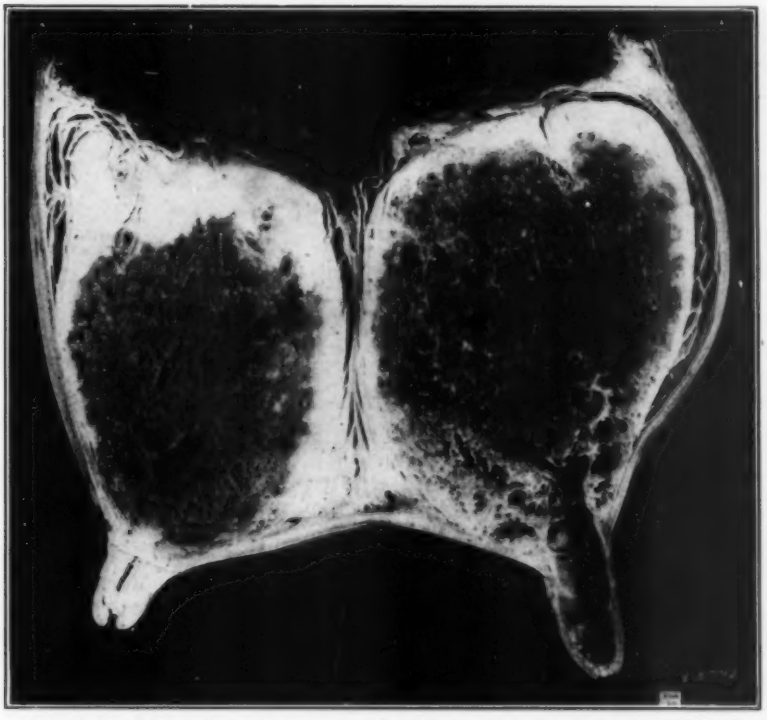

FIG. 4A. This photograph shows the destructive effects of active calfhood infection with pus discharge. The affected front quarter (at the left) resulted from an infection of the quarter at the age of one month. Note the fibrous infiltration in the teat and adjacent area. Rear quarters are shown in Fig. 4B.

FIG. 4B. B, shows the structure of the rear quarters. The cause of the loss of the left rear quarter is not known.

that had invaded the left rear quarter territory. In the right front quarter, which appears at the left in figure 4A, the gland tissue appears to be more nearly normal, although actually it was woody. It is obvious that communication between the gland tissue and the teat canal in both the right front and left rear quarters had been cut off by the formation of fibrous or scar tissue. In all probability the gland tissue made a fairly complete development and secreted milk at calving time, but involution resulted soon thereafter because of inability to remove the milk.

The mammary gland development of No. 859 was not examined until she was 3 months of age. At that time the left rear quarter was definitely re-

tarded and tubular in form. At 3 months 26 days the development was irregular. Subsequently up to 18 months there was nothing to suggest irregularity in that quarter. However, it differed somewhat from the others before calving. Milk was never obtained from the left rear quarter and soon after calving the quarter was obviously deficient. There is no record of infection or injury in this quarter although either could have occurred without detection prior to the first examination. The cow produced liberally despite the complete failure of the left rear quarter in milk secretion. The cow's record for 365 days at 6 years 2 months was 20,163 pounds of milk and 668 pounds of butterfat. Photographs of the gross anatomy of this udder are not yet available.

No. 882 was not examined for mammary gland development until the age of 4 months. At that time the right rear quarter hung lower than the others. At the age of 4 months 28 days the right rear quarter contained a long tubular formation "like a nail." A heavy fibrous growth running the entire length of the teat was noted at the 6-months examination. Nothing indicative of abnormality was recorded subsequently until shortly before calving when an unusually small opening from the teat to the cistern was noted. This quarter was non-functioning from the time of first calving. Soon the quarter became relatively light and stringy and was obviously "blind." Her production at 3 years 2 months was 11,919.9 pounds of milk and 439 pounds of butterfat. The udder was destroyed at the time of slaughter.

No. 881 was examined regularly beginning at the age of 13 days. At 4 months 2 days the left rear quarter was greatly enlarged, hard and discharging pus from the teat. The cause of the disturbance was not known. At the 5-months examination the same quarter contained a hard core that extended into the teat and at the 6-months observation the condition was essentially the same. At the 12-months examination the left rear quarter contained a distinct tubular shaped body of fiber that extended its entire depth. Before calving this quarter became enlarged, and intensely swollen. Following calving the quarter failed to yield milk. At the first examination after calving it was described as "blind but oversized." The cow's 365-day production commencing at 2 years 12 days was 9,391 pounds of milk and 341 pounds of butterfat. It is noteworthy that in this case, like that of 1245 the affected quarter became larger than the others before calving. Photographs of the gross structure of this quarter are not available but it appears likely that in this case also involution resulted from some obstruction that prevented removing the secreted milk.

The mammary gland development of No. 1001 was well above the average. No disturbance or abnormality was noted until the 5-months examination when the left front quarter was greatly enlarged, with the teat swollen and more than twice the length of the others. At six months it appeared that the disturbance had largely disappeared. Although there was no definite infor-

mation to substantiate it the abnormality was attributed to sucking. No irregularity in this quarter was noted at 9, 12 or 18 months or at any of the four pre-calving examinations. The left front quarter was "blind" from the beginning of the first lactation. She produced 6,988 pounds of milk and 430 pounds of butterfat in 365 days, commencing at 2 years 1 month of age. When sections were removed for histological study after slaughter a hard lump was noted in the upper part of the left front quarter. Photographs of the gross structure of the quarter were not made.

Up to the age of four months the mammary gland development of No. 695 appeared to be uniform and above the average for the breed. At 5 months the right rear quarter was undersized, but no explanation was noted. At 6 months the left rear quarter was badly swollen, the teat was enlarged and was discharging pus. At 9, 12 and 18 months the development appeared to be uniform and above the average. Before calving there was no marked tendency for the left rear quarter to differ from the corresponding one on the right. However, it was "blind" from the beginning of the first lactation and soon became very small, fibrous and stringy. Production amounted to 9,756 pounds of milk and 581 pounds of butterfat at the age of 2 years 7 months. Photograph of the structure of this quarter is not available.

There was nothing notably unusual about the mammary gland development of No. 660 in its early stages. It appeared to be typical in form at 5 months and regular but slightly retarded at 9 months. At 12 months, however, the left front quarter consisted chiefly of a hard ball of fibrous tissue and the rear quarters were enlarged, ball-shaped and flabby. The udder showed indications of having been sucked. Subsequently abnormalities were not recorded until at an examination 2 days after first calving there was a "swelling in the teat" of the left front quarter. At a number of periodic examinations this same quarter was described in such a way as to indicate it was definitely abnormal and six months after calving it was described as "blind." The records do not show definitely whether the left front quarter failed entirely to function or whether it became blind soon after first calving. This quarter apparently did not function at any subsequent lactation. Apparently its failure resulted from the disturbance noted at the age of 12 months and there is reason to believe that sucking was the cause. Despite the possible handicap resulting from the disturbance noted she produced 12,885 pounds of milk and 678 pounds of butterfat in one year at the age of 7 years 1 month. The left quarters were used for histology studies and a photograph of their gross anatomy was not obtained.

The mammary gland development of No. 1428 was examined regularly beginning at 18 days of age. Grades for relative degree of development were high at each examination from 1 month to 12 months and no significant abnormality was noted. At the 18-months examination the left rear quarter was greatly enlarged and discharging an abundant quantity of thick, bright

yellow pus. Although bred 17 times she failed to conceive and was slaughtered at the age of 2 years, 5 months, 22 days. Before slaughter the left rear quarter of the udder was greatly enlarged and apparently full of fluid. This fluid amounted to 2.75 pounds and resembled milk. About 0.35 pound of colostrum-like material was removed from the right rear quarter. After the contents were removed the rear quarters were found to be fibrous and stringy in character. Despite her failure to conceive the gross appearance of the mammary gland tissue in the rear quarters, in which lactation apparently was abnormally stimulated in some unknown manner, closely resembled

FIG. 5A. This photograph shows the structure of the two rear quarters of an udder in which the left rear quarter discharged pus copiously at 18 months of age. Fluid was found in both rear quarters when this heifer was slaughtered despite the fact she was a non-breeder.

that of a lactating cow. The two front quarters, however, consisted to a much greater extent of scattered areas of gland tissue imbedded in fat, a condition essentially typical of that generally found in heifers and cows that never conceived. Figure 5A shows the type of cisterns in the rear quarters and 5B shows the general structure of the front quarters.

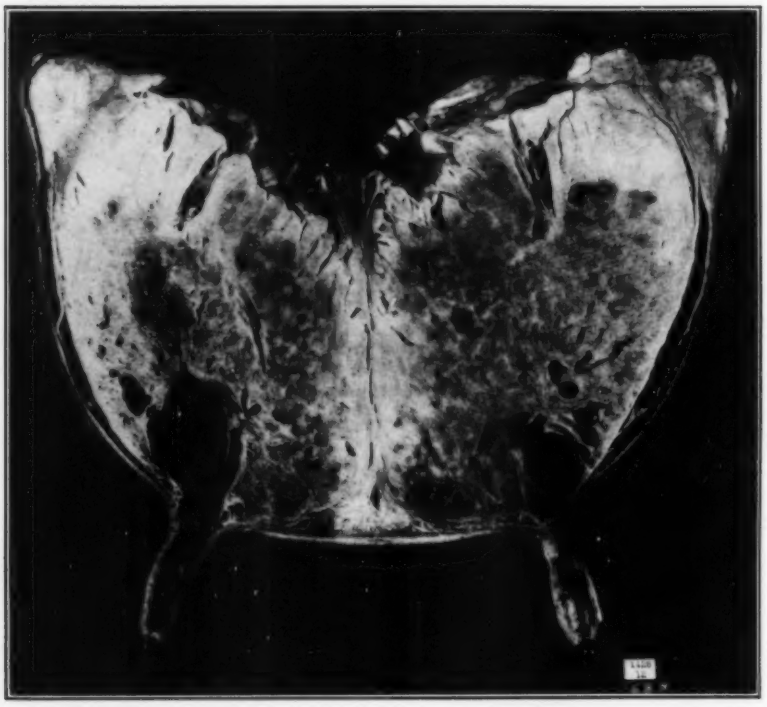

FIG. 5B. The mammary gland structure of the front quarters (rear quarters shown in Fig. 5A) which is fairly typical for a non-breeding animal that never conceived.

There was some indication of a retarded development in the right rear quarter of the udder of No. 1412 at the early age of 25 days, and at subsequent monthly examinations up to 5 months of age the right side seemed to be slightly smaller than the left. At 6 and 9 months no abnormality was noted and the development was above the breed average. At 12 months the right rear quarter seemed a trifle larger than the left rear but at 18 months no difference was recorded. The outstanding point about this case was a pus discharge from the right rear quarter approximately 3½ months before calving. The cause of this disturbance is not known. Two months after calving an indication of a deficiency in the size of the right rear quarter was noted.

At subsequent examinations the "lightness" of this quarter was consistently noted and the deficiency seemed to be progressive. Her record as a 2-year-old amounted to 10,071 pounds of milk and 625 pounds of butterfat. Approximately 4 months after second calving the milk yield from separate quarters was measured and it was found that only 10.3 per cent of her total milk yield was secreted by the right rear quarter when normally approximately 30 per cent would be expected. Here is a case where the infected quarter did not become blind but its functioning ability was greatly impaired.

No. 1438 showed no irregularity at any examination until at the examination made 20 days before calving, the right rear quarter was hard and lighter than any of the others and it was noted at the time that the quarter probably was "bad." On the day following calving it was found to be "blind," and 23 days later the same quarter was discharging pus. Figure 6 shows the appearance of the tissue in both rear quarters. Small areas of fairly normal appearing tissue may be noted in the nonfunctioning right rear quarter but connection between them and the cistern or the teat canal seems to have been destroyed so that milk, although present, could not be removed.

A more recent case is that of cow No. 1488. Her mammary gland development was better than average and fairly uniform until 18 months of age when the right front teat was described as higher than the others. At the examinations made 25 and 12 days before calving, the right front quarter was swollen, hard, and woody with a thickened condition in the teat. Milk has not been obtained from this quarter at any time.

A case of considerable interest is that of No. A 134. This heifer had a greatly enlarged udder at the age of 12 months when three of the quarters were filled out and discharging a milk-like fluid. Twelve days later the condition was even more extreme and the udder was very much distended. She was milked for about 2½ weeks after the appearance of fluid in the udder but the quantity of fluid obtained was not recorded. At the age of 21 months the three quarters were long and tubular, and apparently still contained milk. The udder was of extremely poor shape. At approximately 2 years of age the condition was more pronounced. The right front quarter was about 8½ inches deep and 3 inches in diameter and tapered into the teat. The right rear quarter was still longer and larger in diameter. The left front quarter was about 5½ inches long and 2¼ inches in diameter. The left rear quarter was almost entirely undeveloped. The enlarged quarters were cylindrical in shape except for tapering toward the teat. Each of the two right quarters appeared to contain an extremely large cistern but little glandular tissue. The left front quarter was more firm, harsh, and fibrous and its cistern appeared to be filled with a soft rubber-like substance. The cistern in the left rear quarter was very small and not readily detectable. She proved to be a non-breeder, and was slaughtered at about 24½ months of age. Her vaginal tract was found to be small and shortened, the uterus was in-

FIG. 6. Structure of the two rear quarters of an udder in which an abnormality was first noted 20 days before calving. The quarter on the right was "blind" from the time of first calving. Note the occlusion in the region of the cistern.

fantile, the ovaries were definitely abnormal, and one of the Fallopian tubes was occluded. The watery fluid milked from the two right quarters immediately before she was killed amounted to 3.47 pounds. Nothing could be obtained from either of the left quarters. The amounts of formalin injected into the four quarters were: right front, 1277 cc., right rear 2426 cc., left front, 59 cc., and left rear 59 cc. The formalin did not appear to penetrate far beyond the cistern in either of the two left quarters. The gross structure

FIG. 7. Structure of tissue in the rear quarters of an udder in which the right rear quarter and both front quarters became distended at 12 months of age and remained so until time of slaughter at 24½ months. Note the enormous cistern in the affected quarter, a condition which is often found in cases of precocious activity of mammary glands.

of the rear quarters of this udder is shown in figure 7. It is suspected that this udder was sucked previous to its abnormal development at about 12 months of age. Nothing is known, of course, as to what the condition of the udder would have been after calving if this animal had been a normal breeder.

The cases described give some idea of the probable results to be expected from injury, infection or abnormal stimulation of the udder in heifers. Numerous cases of blind and deficient quarters of the udders have been observed following calving in cases for which there was no recorded history of disturbance in the calf. However, in every instance on record where pus discharge from the teat has occurred in the calf or young heifer, the affected quarter has failed to function from the beginning of lactation, except that of No. 1412, where the pus discharge occurred during late pregnancy and in that case the quarter became deficient after parturition. Pus discharge was not noted in the case of the non-breeding heifer No. A 134 but the appearance of the udder indicates that it was definitely abnormal and strongly suggests that its producing capacity would have been affected.

In nearly every case where udder infection has been found in the calf or young heifer, the affected quarters were abnormal in development for a time, but filled out approximately the same as the unaffected quarters before calving time. After calving, however, milk was not obtainable from these infected quarters and a marked decrease in size soon followed. In some of these cases, where post-mortem examinations were made, it appeared that there had been a development of secretory tissue and a development of fibrous or scar tissue in the region of the cistern. These findings make it appear probable that an early infection or injury does not necessarily cause a cessation in the development of the gland tissue in the affected quarter; that milk may be secreted in such quarters at calving time; and that occlusion of the outlet prevents removal of the milk thereby resulting in the involution of the tissue.

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COMPARATIVE VALUE OF NAPIER AND SUDAN GRASS WHEN USED AS SOILING CROPS FOR DAIRY COWS

L. A. HENKE[1] AND G. W. H. GOO[2]
The University of Hawaii, Honolulu

In areas adjacent to Honolulu and to other cities in the Territory, dairy cattle are commonly kept in small paddocks, although in regions remote from the cities, they are grazed the year round. In the paddocks the cattle receive their roughage from green soiling crops in racks. Concentrates are fed in the milking barn at the time of milking. The use of all available areas for growing soiling crops rather than for pasture is necessitated by the scarcity of the land.

Among various grasses grown in Hawaii as soiling crops, Napier grass (*Pennisetum purpureum*) and Sudan grass (*Sorghum vulgare sudanense*) occupy prominent places. The purpose of the investigation reported herein was to determine the relative value of these two grasses when grown as soiling crops for milking cows.

PLAN OF EXPERIMENTS

One general plan was followed in each of four experiments, with six cows selected for three experiments and eight cows for one experiment. The animals for each experiment were divided into two lots as comparable as possible with regard to milk production, body weight, age, days since calving, and date again due to freshen.

The reversal system of feeding was followed, and each trial was limited to 12 weeks to avoid the effects of advancing pregnancy. This reversal arrangement is clearly shown in table 1.

TABLE 1

Reversal system of feeding soiling crops

	Soiling crop fed	
	Lot I	Lot II
First 4 weeks	Sudan grass	Napier grass
Second 4 weeks	Napier grass	Sudan grass
Third 4 weeks	Sudan grass	Napier grass

The first week of each 4-week period was designed to accustom the cows to change of feed and results of only the last 3 weeks of each feeding period

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[1] Animal Husbandman, Hawaii Agricultural Experiment Station.

[2] Assistant in Animal Husbandry (resigned Nov., 1938), Hawaii Agricultural Experiment Station.

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were used in computations. The averages of the first and third periods of each lot were compared with the second or middle period, thus compensating for decreasing production due to advancing lactation. The results reported for each experiment are again the averages of lots I and II, thus compensating for conditions which could not be controlled, such as temperature, rainfall, *et cetera*.

Ample protein and total nutrients based on the Morrison Standard were available for the cows in both lots in each of the four experiments regardless whether Sudan or Napier grass was fed. However, because of the higher consumption of Sudan grass and its higher protein content, protein intake was in greater excess of requirements when Sudan rather than Napier grass was fed.

Both lots of cows in each experiment were kept in small paddocks, except during milking hours. The roughages were fed in racks, and roughages unconsumed after 24 hours were weighed back. Concentrates were fed in the barn at milking time, twice daily. In each experiment all cows received the same concentrate mixture, but in quantities adjusted to the milk production of each cow during the first and second 4-week periods of each trial. During the last 4-week period, the quantity of concentrates fed each cow was adjusted so that the average consumption of the first and third 4-week period would equal that of the second 4-week period. Thus, a cow fed 15 and 14 pounds of concentrates daily during the first two periods, respectively, would be fed 13 pounds daily during the last period to avoid a variable other than roughage. The quantity of concentrates fed the third period

TABLE 2

Milk production, butter fat content, and weights of cows fed Sudan and Napier grasses

	Average daily milk production	Average butter fat content of milk	Average concentrates fed per day	Average green roughage consumed per day	Average live weight of cows
	Pounds	*Per cent*	*Pounds*	*Pounds*	*Pounds*
Sudan grass					
Experiment I	29.7	3.55	15.8	68.7*	996
Experiment II	31.0	3.38	16.6	58.5*	1,012
Experiment III	24.4	4.24	16.2	48.1†	997
Experiment IV	25.5	4.18	14.2	57.7†	1,018
Average	27.6	3.84	15.7	58.2	1,006
Napier grass					
Experiment I	27.8	3.68	15.8	63.5*	999
Experiment II	28.5	3.44	16.6	58.0*	1,008
Experiment III	22.3	4.38	16.2	45.0†	965
Experiment IV	22.9	4.31	14.1	50.9†	1,011
Average	25.4	3.95	15.7	54.3	996

* Roughages fed whole—only the finer parts of plant consumed.
† Roughages cut in ensilage cutter before feeding.

was generally in rather close accord with the needs of the cow based on her milk production.

Live weights of each cow were taken at the end of each week and averaged; butter-fat percentages were based on composite samples (four milkings), also taken at the end of each week for each cow. The summarized results are shown in table 2.

In each experiment milk production was higher when Sudan grass was fed, the difference averaging 8.66 per cent. While this is not a large difference, it was a consistent one, only 2 of the 26 cows in the 4 experiments producing more milk when Napier grass was fed.

Statistical analysis[3] consisted of calculating the differences between average daily milk productions of the individual cows when fed Sudan grass and when fed Napier grass, and determining the significance of the mean difference. The statistics are summarized below:

n = number of comparisons between Napier and Sudan grasses = 26

Sx^2 = sum of squares of deviations from the mean difference in production = 45.33

m = mean difference in production = 2.3 pounds

$$Sm = \text{standard error of the mean difference} = \sqrt{\frac{Sx^2}{n(n-1)}} = \sqrt{\frac{45.33}{26(26-1)}}$$

$$= \pm 0.26$$

$$t = \frac{m}{Sm} = \frac{2.3}{0.26} = 8.8$$

For a probability of 1 per cent with 25 degrees of freedom, a t value of 2.7 is necessary. The t value calculated above greatly exceeds the required amount, indicating that the difference in production due to feeding of Sudan grass was highly significant.

Percentages of butter fat in the milk averaged higher in each trial when Napier grass was fed, 19 of the 26 cows showing a gain in butter-fat content on this grass. The mean gain was 0.11 per cent, with a standard error of 0.0453. The calculated t value was 2.4 which indicated significance at the 5 per cent probability level.

The differences in live weight of the cows were not significant.

Consumption of green roughages as shown in table 2 was higher on Sudan grass, indicating that the greater value of Sudan grass for milk production was probably due to its greater palatability, resulting in larger amounts consumed.

In the first two experiments the green roughages were fed whole, immediately after cutting, and the portions weighed back after 24 hours consisted largely of the coarser parts of the stems. In the last two experiments

[3] Snedecor, G. W. Statistical Methods, 341 pp., illus. Ames, Iowa, 1937. The help of Dr. J. H. Bartlett of the Hawaii Station in the statistical analysis of the data is gratefully acknowledged.

the green roughages were put through an ensilage cutter prior to feeding. Samples were taken daily of the green feed as well as the portions unconsumed after 24 hours. Moisture determinations were made on all these daily samples, and composites of each 3-week period were analyzed for crude protein, ether extract, nitrogen-free extract, fiber, and ash, thus making it possible to determine actual nutrient intake in the roughages consumed. These data are shown in tables 3 and 4.

TABLE 3

Composition of Sudan and Napier grasses (calculated to a water-free basis)

	Crude protein	Ether extract	Nitrogen-free extract	Crude fiber	Ash	Total nutrients
	Per cent	*Per cent*	*Per cent*	*Per cent*	*Per cent*	*Per cent*
Sudan grass						
Experiment III	6.16	1.30	45.23	37.28	10.02	91.58
Experiment IV	8.37	1.13	42.32	36.35	11.83	89.59
Average	7.26	1.21	43.77	36.81	10.92	90.58
Napier grass						
Experiment III	4.76	1.12	40.08	38.86	15.19	86.22
Experiment IV	5.55	1.24	38.54	38.43	16.24	85.32
Average	5.15	1.18	39.31	38.64	15.71	85.77

TABLE 4

Nutrients contained in roughages consumed by cows in Napier-Sudan experiments III and IV (calculated to a per-cow-per-day basis)

	Weight of green forage consumed	Dry matter consumed	Total protein consumed	Total nutrients consumed
	Pounds	*Pounds*	*Pounds*	*Pounds*
Sudan grass	52.9	11.5	0.87	10.4
Napier grass	47.9	7.6	0.41	6.5

It appears from the above table that, since concentrates fed the cows were the same (see table 2), the lower milk production on Napier grass was due to the lower palatability and consequent lower protein and total nutrient intake.

SUMMARY

In four experiments comparing green Sudan with green Napier grass as a soiling crop fed to dairy cows, the former proved superior in each test on the basis of quantity of milk produced. Milk production averaged 8.7 per cent higher, and roughage consumption 7.2 per cent higher when Sudan grass was fed. The higher production on Sudan grass resulted from a greater nutrient intake because of greater palatability and higher nutritive value. Butter-fat tests were slightly higher when Napier grass was fed. Analyses of the grasses show that the Sudan grass was higher in protein, nitrogen-free extract, and total nutrients than Napier grass.

THE LIPIDS OF MILK. II. THE FAT ACIDS OF THE SPHINGOMYELIN–CEREBROSIDE FRACTION

FLOYD E. KURTZ AND GEORGE E. HOLM

Division of Dairy Research Laboratories, Bureau of Dairy Industry, U. S. Department of Agriculture

Phospholipids occur in all cells and to the extent of approximately 0.034 per cent in the milk of cows. Though their exact function in the animal body is not known, they have been thought to be intimately associated with fat synthesis. Although a minor constituent of food they may prove to be of especial significance.

Osborne and Wakeman (1) identified a monoamino- and, presumably, a diamino-phospholipid in milk, and their data indicated that oleic and stearic acids were constituents of the monoamino fraction.

Sasaki and Hiratsuka (2) concluded that the lecithin in milk contains myristic and lauric acids; and the cephalin of milk, palmitic and lauric acids.

Kurtz, Jamieson, and Holm (3) isolated the phospholipids from sweet cream buttermilk powder and separated them, on the basis of their solubilities in ether, into two distinct fractions: an ether-soluble lecithin-cephalin fraction, and an ether-insoluble sphingomyelin-cerebroside fraction. Their analysis of the lecithin-cephalin fraction showed that the fat acids consist of 5.2 per cent myristic, 16.1 per cent stearic, 1.8 per cent arachidic, 70.6 per cent oleic and 6.3 per cent of what was presumably a dicostetrenoic acid.

The analysis of the sphingomyelin-cerebroside fraction is presented in this paper.

EXPERIMENTAL

Isolation of material

The isolation of the phospholipids has been described fully in the first paper of this series (3). It consisted essentially in the extraction of three hundred pounds of sweet cream buttermilk powder with a mixture of 3 parts of ethanol and 1 part of gasoline. The extract was concentrated in a vacuum pan and then added to acetone. The precipitate which formed was freed from fat by a procedure involving repeated precipitations with acetone from ether solutions and from water emulsions and, finally, trituration of the precipitate with hot acetone.

During the purification of the phospholipids, as outlined above, it was observed that if an ether solution of the phospholipids was kept over night at 0° C., a light brown precipitate was formed. It was found that the solubility of this precipitate in the ether solution of lipids decreased as the lipids became more pure. Consequently, additional precipitates were obtained, at

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each stage in the purification, by holding ether solutions of the phospholipids at 0° C.

The ether-soluble lecithin-cephalin fraction weighed 580 grams. The ether-insoluble fractions were combined and together weighed 90 grams. This material was freed from contaminating lecithin and cephalin by crystallization from ethanol followed by washing with ether. This procedure was repeated until the originally discolored sticky mass became a white powder. The yield was 60 grams. The absence from its hydrolytic products of oleic acid, which forms 70 per cent of the acid constituents of the lecithin-cephalin fraction, indicates that it had been satisfactorily freed of these two phospholipids.

The material gave a positive orcinol test for carbohydrate. It contained 2.52 per cent nitrogen and 2.06 per cent phosphorus. These values are consistent with those for a mixture of sphingomyelin and cerebrosides in approximately equal amounts. When rubbed with concentrated sulphuric acid a deep purple color was produced. Thudichum (8) found that this reaction takes place when sphingosine and a sugar are treated with concentrated sulphuric acid. The material was practically insoluble in ether and in petroleum ether. Its solubility in 95 per cent ethanol at room temperature was 2 per cent; in hot ethanol, 13 per cent; and in chloroform at room temperature, 20 per cent.

Acids of the sphingomyelin-cerebroside fraction

Fifty-five grams of the material was hydrolyzed in methanol containing 10 per cent by weight of sulphuric acid, according to the method of Merz (4). Of the fat acid esters formed, 15.2 grams separated from solution, on cooling. These were saponified with alcoholic potassium hydroxide, diluted with water, and extracted with ether to remove unsaponifiable material. The yield of acids was 13.4 grams.

The filtrate containing the soluble methyl esters was diluted with water and extracted with ether. The concentrated extract was saponified, extracted with ether to remove unsaponifiable material, acidified, and the acids recovered. The yield was 5.6 grams of acids.

Analysis of these acid fractions gave the following values:

	Fraction A *Acids from insoluble methyl esters*	*Fraction B* *Acids from soluble methyl esters*
Melting point (without recrystallization)	76° C.	Sticky semi-solid
Mol. Wt. (by saponification)	355	345
Iodine No. (Hanus)	0.17	5.4
Acetyl No.	10.4	—

Fraction A—acids from insoluble methyl esters

Fraction A was esterified with ethanol and fractionally distilled from a Ladenburg flask.

TABLE 1
Data on distillation of esters of Fraction A

Fraction	Pressure	Temperature	M.P. of esters	M. Mol. Wt. of esters	Wt. of fraction
No.	*mm.*	*°C.*	*°C.*		*Grams*
A-1	1.0	165–200	50.0	380.0	4.67
A-2	1.0	200–205	51.2	384.0	7.68
A-residue	—	205–210	—	397.0	0.75

By repeated recrystallization of the acids of Fraction A-2, 1.5 grams of lignoceric acid[1] was isolated. It had a m.p. of 80° C. and a molecular weight of 368.4 (Theor. 368.38). About 0.2 gram of lignoceric acid was also isolated from the residue. The isolation of this acid from Fraction A-1 and its purification was attained only with great difficulty. The acetyl value indicated the presence of one or more hydroxy acids. Presumably the presence of such acids was the source of the difficulty experienced in purifying the lignoceric acid. The residues were combined, re-esterified and finally redistilled and the acids isolated. Attempts at isolation from this fraction of acids of lower molecular weight than that of lignoceric acid resulted each time only in the final isolation of material consisting entirely or mostly of lignoceric acid.

Assuming that the acids present in the fractions analyzed have an average molecular weight equal to that of stearic acid, a calculation based upon the molecular weight data obtained on each of the fractions indicates that the material of Fraction A consists of approximately 80 per cent of lignoceric acid.

Fraction B—acids from soluble methyl esters

After removal of the bulk of the lignoceric acid from the acids of this fraction by crystallization from ethanol the remaining acids were converted into lead salts and extracted with ether to separate the saturated and unsaturated acids. From the ether-insoluble lead salts 1.0 gram of acids and from the ether-soluble lead salts 0.25 gm. of acids was recovered. The following analytical values were obtained:

	Fraction B-1 *Acids from ether-insoluble lead salts*	*Fraction B-2* *Acids from ether-soluble lead salts*
Mol. wt.	303	369.7
Acetyl value	7.8	19.6
Iodine No. (Hanus)	3.7	28.3
Yield in grams	1.0	0.25

The acids from the ether-insoluble lead salts (Fraction B-1) were frac-

[1] Jantzen and Tiedcke (5) found that the lignoceric acid obtained from peanut oil is a mixture. Chibnall, Piper, and Williams (6) found that the lignoceric acid obtained from brain cerebrosides also is a mixture. They thought that its most probable composition is: 10 per cent hexacosanoic, 80 per cent tetracosanoic, and 10 per cent docosanoic acids.

tionated by distillation under reduced pressure with the aid of an electrically heated semi-micro fractionating column. The data are given in table 2.

TABLE 2

Data on distillation of acids from ether-insoluble lead salts

Fraction	Pressure	Temperature	M. Mol. Wt. (by Sapn.)	Wt. of fraction
No.	*mm.*	*°C.*		*grams*
1	1.5	–190°	259.6	.35
2	1.5	190–205°	300.7	.13
3	2.0	230–232°	331.3	.20
Residue	....	.....	330.7	.20

From the mean molecular weight of the first fraction (259.6) it was evident that one component had a molecular weight at least as low as that of myristic acid. The first as well as the second fraction was subjected to steam distillation and a product was finally isolated which had a molecular weight of 228.8, or that of myristic acid. Other products were isolated from the steam distillates, varying in their mean molecular weight from 240 to 300, but in all cases the analytical values were such as to indicate mixtures. The values in table 2, indicate that at least 50 per cent of the acids in this fraction are of a molecular weight greater than 300.

The acids from the ether-soluble lead salts (Fraction B-2) after treatment with norite were obtained as a light brown viscous liquid. Their mean molecular weight (369.7) is that of an acid of 24 carbon atoms. Because of the extremely small amount of material available, the nature of the acids of this fraction can only be conjectured. The analytical values do, however, indicate that these acids are of types similar to those found from other analogous sources of lipids. The acetyl value indicates a hydroxy acid, and the iodine value an unsaturated acid. In view of the fact that a saturated hydroxy acid of so high a molecular weight would probably form an ether-insoluble lead salt, it is likely that the hydroxy acid of this fraction is also unsaturated. Klenk (7) has isolated such an acid, an α hydroxy tetracosenoic acid, from the cerebrosides of the brain. The iodine number is sufficiently high to account for, in addition, a non-hydroxy unsaturated acid. Nervonic acid, a mono unsaturated acid of 24 carbon atoms is also characteristic of brain cerebrosides.

SUMMARY

The acetone-insoluble, ether-insoluble lipids of milk were isolated and found to consist of approximately equal parts of sphingomyelin and cerebrosides. Their amount was approximately 15 per cent of that of the ether soluble lecithin-cephalin material formerly isolated and analyzed.

Their fat acid constituents were analyzed and found to consist chiefly of lignoceric acid (approximately 80 per cent) which was isolated and iden-

tified. The other 20 per cent of acids was found to be a very complex mixture of saturated, unsaturated, hydroxy saturated, and presumably hydroxy unsaturated acids, no one with the exception of myristic acid being present in sufficiently great concentration to make isolation possible with the limited supply of available material. The indications were that myristic acid is present in a concentration of less than 1 per cent. The presence of lignoceric acid in high concentration and the indicated nature of the other acids are in harmony with the nature of the acids of sphingomyelin and cerebrosides obtained from other sources.

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COOKED AND OXIDIZED FLAVORS OF MILK AS AFFECTED BY FERROUS IRON*

I. A. GOULD

Department of Dairying, Michigan State College, East Lansing, Michigan

INTRODUCTION

In a previous study (1) data were presented showing the correlation between cooked and oxidized flavors of milk, the cooked flavor being differentiated from the "pasteurized" or "heated" flavor which frequently results from ordinary pasteurization procedures. A relationship was found to exist between copper-induced oxidized flavor and the cooked flavor when the copper was added before heating the milk, whereas there was no such relationship noted when the copper was added subsequent to heating. In the former case, the cooked flavor usually appeared at temperatures of 84–86° C., whereas temperatures of 76–78° C. sufficed to cause the flavor when the copper was added following the heat treatment. Coincidental with the appearance of the cooked flavor when the copper was added before heating was the failure of the milk to develop the oxidized flavor, *i.e.*, the milk heated to 84–86° C. or above did not become oxidized whereas that heated to lower temperatures developed this flavor defect. Such a relationship between the cooked and oxidized flavors did not exist when the copper was added following the heating and all of the samples became oxidized even though heated to 90° C.

Further, this study showed hydrogen sulphide liberation to be correlated with the cooked flavor and to be responsible for the heat retardation of the development of the oxidized flavor. Copper and ferrous iron both were found to have a retarding influence on the cooked flavor, with the copper being decidedly more effective than iron in this respect. The difference between these metals in this connection was ascribed as being due perhaps to the greater affinity of copper for the sulphides which were formed during the heat treatment. For a fuller explanation of the problem and for a complete literature review the reader is referred to the previous publication (1).

Since the earlier work in this connection dealt chiefly with the relation of cooked and oxidized flavors when copper served as the catalyst for inducing the oxidized flavor, it was thought desirable to ascertain if ferrous iron would give similar results. Consequently, the study reported in this paper was conducted to determine (a) the relation between the cooked and iron-induced oxidized flavors when the iron is added before and after heating the milk, (b) the possible correlation of the findings with the liberation of the milk sulphides.

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EXPERIMENTAL

Procedure: The milk used in this study was from the mixed herd supply of the College creamery. The heating trials were conducted using a 5-liter round bottom flask suspended in a boiling water bath. An electrically driven glass-rod agitator stirred the milk during its heating period. A bent glass tube was used as a siphon by which samples were taken for flavor examinations. An accurate thermometer was suspended in the milk during its heat treatment.

Approximately 3.5 liters of milk were heated in one trial. The milk was raised to the desired temperature in about 15 minutes. The temperatures at which the samples were taken were accurate within $\pm 0.5°$ C. The samples were collected in glass sample jars and were immediately cooled by placing the jars in cold water.

Cooked flavor determinations were made within 30 minutes following the heat treatment, and the milk was again examined both for cooked and oxidized flavors after 48 hours storage at 40° F. The samples were keyed in such a manner that their identity remained unknown to the judges. Flavor examinations were made usually by two experienced milk judges.

The oxidized flavor was developed by the addition of a ferrous chloride solution added at a rate of 2.8 p.p.m. of the iron. The iron was added either immediately before or immediately after the milk had been heated.

The sulphide determinations were conducted as described previously (1). In brief, the method consists of heating and aerating 4.5 liters of the milk and drawing the vapors through a filter paper dampened with lead acetate solution. The darkening of the paper due to the formation of lead sulphide indicates the liberation of hydrogen sulphide from the milk.

Results: Results secured when the iron-contaminated milk was heated are illustrated in figure 1. The figure represents the results from five different

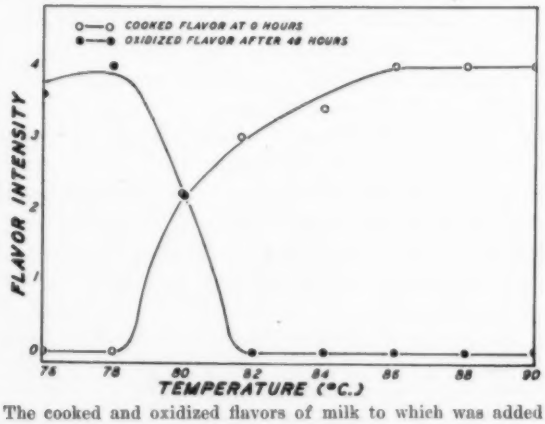

FIG. 1. The cooked and oxidized flavors of milk to which was added 2.8 p.p.m. of ferrous iron before heating.

trials, all of which gave practically identical results. The intensity of the flavor was graded as follows: 1 = questionable, 2 = faint, 3 = distinct, and 4 = strong.

The graph shows a definite relation between the cooked and oxidized flavors. Temperatures of 80–82° C. or higher usually produced the cooked flavor, whereas the lower temperatures did not. These cooked flavor results were in general unchanged whether the flavor examination was made immediately after heating or after the 48-hour storage period. This is a point of difference between these findings and those reported earlier for copper (1). The oxidized flavor usually developed in those samples which were heated to 80° C. or lower, whereas the flavor failed to develop in those samples subjected to higher temperatures. In other words, samples which possessed the cooked flavor did not become oxidized, and vice versa. Exceptions occurred in the milk heated to 80° C. which at times was criticized as being cooked and also developing an oxidized flavor. This temperature, *i.e.*, 80° C., might be said to be the critical temperature in this connection and is the dividing point between the oxidized and cooked flavors. These results are similar to those reported earlier for copper with the exception that the critical temperature for copper was found to be approximately 84–86° C.

Several trials were also conducted to determine the effect on the cooked and oxidized flavors of milk of adding the ferrous iron after heating. These results are portrayed in figure 2, and in general are not greatly different

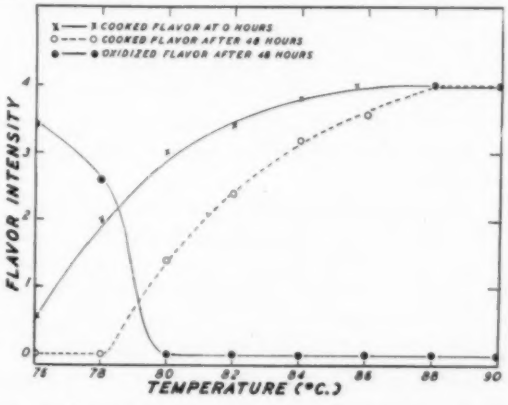

FIG. 2. The cooked and oxidized flavors of milk to which was added 2.8 p.p.m. of ferrous iron after heating.

from those illustrated by figure 1. This graph, however, does show the influence of the storage period upon the cooked flavor. The cooked flavor of the milk, as determined immediately after the heat treatment, occurred at the normal temperature of 76–78° C. Following the addition of the iron

and after the 48-hour period, the lowest temperature at which the cooked flavor was detected was 80° C. As in the case when the iron was added before heating approximately 80° C. was found to be the critical temperature. However, there seemed to be a slight tendency for this temperature to be shifted to a lower point although the difference appears negligible. The graph shows that the milk heated to 76° or 78° C. developed an oxidized flavor, whereas this flavor did not develop in the milk heated to 80° C. or above. Conversely, the cooked flavor was present in the milk heated to 80° C. or above even after 48 hours of storage. The cooked flavor observed immediately after heating in those samples heated to 76° or 78° C. had disappeared during storage, being replaced by the oxidized flavor.

Figures 1 and 2 show that the influence of heat in preventing the development of the oxidized flavor as induced by ferrous iron is equally as effective irrespective as to whether the iron was added before or after heating. This indicates a difference in the influence of heat on the oxidized flavor caused by iron or copper, inasmuch as the previous study (1) showed the copper-induced flavor to be greatly affected by the period of contamination, viz., whether before or after heating. Further, the fact that the cooked flavor of the milk persisted following storage irrespective as to whether the iron was added before or after the heating process, indicates another difference between the action of iron and copper in regard to this flavor.

To verify the difference between copper and iron in affecting the disappearance of the cooked flavor, samples of milk were heated to various temperatures and following cooling were divided into three lots. Lot 1 served as the control, Lot 2 contained 2.8 p.p.m. of added ferrous iron, and Lot 3 contained 1.4 p.p.m. of added copper. The samples were stored at approximately 4° C. and were examined for cooked flavors after 0, 6, 24, and 48 hours. The results are presented in table 1, and represent the average of five trials.

These data show the ferrous iron to have no appreciable effect on the persistence of the cooked flavor when the milk had been heated to 82° C. or above. Generally, the cooked flavors of these samples were equally as intense as in the control even after the 48-hour period. However, when the milk had been heated to lower temperatures, the iron-contaminated samples showed more rapid disappearance of the cooked flavor than was observed in the control, although the difference was not great. The samples containing iron which had been heated to 76°–78° C. usually showed oxidized rather than cooked flavors following storage.

On the other hand, the copper had a marked effect on the persistence of the cooked flavor and brought about its total disappearance in all samples by the end of 48 hours. In fact, the flavor had usually disappeared by the end of the 24-hour period, and was usually present after 6 hours only in

TABLE 1

The persistence of the cooked flavor of milk as influenced by the addition of copper or ferrous iron after heating

Temperature of heating	Storage period (hours)											
	0			6			24			48		
	Lot number*			Lot number			Lot number			Lot number		
	1	2	3	1	2	3	1	2	3	1	2	3
°C.												
76	1.2	0.0	0.2	1.0	0.0	0.0	0.0	0.2	0.0	0.0	0.0	0.0
78	3.2	2.8	1.2	3.2	2.3	0.0	2.4	2.2	0.0	0.6	1.0	0.0
80	4.0	3.6	2.4	4.0	3.3	0.0	4.0	3.2	0.0	3.4	2.8	0.0
82	4.0	3.8	3.4	4.0	3.6	0.0	4.0	3.8	0.4	3.8	4.0	0.0
84	4.0	4.0	3.6	4.0	3.8	0.0	4.0	3.8	0.8	3.8	4.0	0.0
86	4.0	4.0	3.8	4.0	4.0	0.8	4.0	4.0	0.8	4.0	4.0	0.0
88	4.0	4.0	3.6	4.0	4.0	2.0	4.0	4.0	1.2	4.0	4.0	0.0
90	4.0	4.0	3.8	4.0	4.0	2.8	4.0	4.0	1.2	4.0	4.0	0.0

* Lot 1 is the control. Lot 2 contains 2.8 p.p.m. added iron, and Lot 3 contains 1.4 p.p.m. added copper. In order to average the results the flavors were scored as follows: 0 = no flavor, 1 = questionable, 2 = faint, 3 = distinct, 4 = strong.

those samples heated to 88° and 90° C. The copper-containing samples were all oxidized by the close of the storage period.

The influence of copper and iron on the persistence of the cooked flavor of milk heated to 80° C. is illustrated by figure 3. This graph shows the

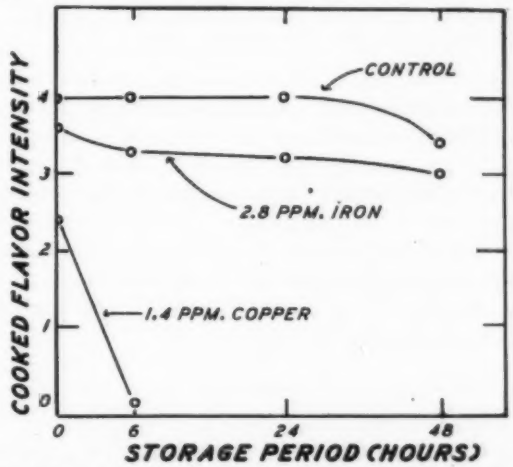

FIG. 3. The influence of copper and iron on the cooked flavor when added to milk which had been heated to 80° C.

marked acceleration of the disappearance of the cooked flavor when copper is added to the milk following the heat treatment, and shows further that ferrous iron has only a small and rather insignificant effect in this connection.

The ability of these metals, *i.e.*, ferrous iron and copper, to affect the liberation of hydrogen sulphide from milk may be correlated directly with their ability to affect the cooked flavor. The influence of these metals on the liberation of the heat labile sulphides of milk as detected by lead acetate paper is shown in table 2.

TABLE 2

*The influence of ferrous iron and copper on the heat labile sulphides of milk**

Treatment of samples	Temperature of heating (°C.)							
	76	78	80	82	84	92	94	98
	Intensity of discoloration of lead acetate paper							
Trial 1								
Milk	0	+	++	–	–	–	–	–
Milk and Fe(ous)	–	0	+	+	++	–	–	–
Milk and Cu	–	–	–	–	–	–	0	0
Trial 2								
Milk	0	0	+	++	–	–	–	–
Milk and Fe(ous)	0	0	+	++	–	–	–	–
Milk and Cu	–	–	–	–	–	0	+	–
Trial 3								
Milk	0	+	++	–	–	–	–	–
Milk and Fe(ous)	0	0	+	++	–	–	–	–
Milk and Cu	–	–	–	–	–	0	0	0

* Iron added at the rate of 2.8 p.p.m. and copper at the rate of 1.4 p.p.m. before heating. Discoloration of lead acetate paper graded as follows: 0 = no discoloration, + = faint discoloration, ++ = distinct coloration (– represents no sample).

These results show that the iron has little retarding influence on the liberation of the sulphides with both the normal milk and the iron-treated milk giving a visible discoloration of the lead acetate paper at temperatures of 78° to 80° C. The normal milk tended to show the discoloration at a slightly lower temperature than the iron containing samples, which is to be expected on the basis of the relationship of the sulphide liberation to the cooked flavor. The copper had a marked influence on the sulphide liberation with no sulphide having been detected in two of the trials even though the milk was heated to 98° C. In one of the trials slight discoloration of the lead acetate disc was noted at 94° C.

DISCUSSION

The data reported herein verify those previously reported (1). The influence of heat on retarding iron-induced oxidized flavor is even more pronounced than in the case of copper, due probably to differences in the combining properties of these metals, either with the liberated sulphide or with the substance furnishing this sulphide, or to differences in oxidizing intensities of the two metals. The almost total lack of combining or oxidizing power by the ferrous iron is reflected by the fact that the oxidized flavor results were practically the same whether the iron was added before

or after the heat treatment. This is further emphasized by the change in the cooked flavors after varying periods of storage.

The close relationship between the cooked and oxidized flavors of milk, and the correspondingly close correlation between the cooked flavor and sulphide liberation, serves to offer further proof that the reducing system which is formed when milk is heated is responsible for the retardation or prevention of oxidized flavor development, rather than that some enzyme is inactivated.

On the basis of these studies, temperatures of 80° C. or above must be used to prevent the oxidation processes which are induced in milk by ferrous iron. Apparently, lower temperatures are not sufficient to create a reducing system capable of hindering these oxidative changes, at least when the iron concentrations are in the range of 1.4 to 2.8 p.p.m.

CONCLUSIONS

The heating of milk momentarily to 80°–82° C. or above prevents the development of an oxidized flavor as induced by 2.8 p.p.m. of ferrous iron. This occurs irrespective as to whether the iron is added before or after the heat treatment.

The appearance of a strong cooked flavor at these temperatures is correlated with the failure of the oxidized flavor to develop. The liberation of sulphides is related to the cooked flavor and prevents the development of the oxidized flavor.

The "critical" temperature is different for milk containing added ferrous iron than for that containing added copper, due to differences in the sulphide combining abilities of these metals, due to differences in their abilities to form complexes with the substance supplying this heat labile sulphur, or due to differences in their oxidizing properties.

ACKNOWLEDGMENTS

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EXPECTATION OF LIFE IN DAIRY COWS*

C. Y. CANNON AND E. N. HANSEN
Iowa State College, Ames, Iowa

Although the expectation of life in humans has been widely investigated and the information obtained about it extensively used by insurance companies and other agencies, there is little information relative to the expected span of life in dairy cattle or other farm animals. Such information on dairy cattle would be useful to the breeder, the buyer and the experimenter, who could then gauge their programs and objectives on something more substantial than guesses.

It is known that replacement rates in dairy herds are rather high, for investigations (1, 2, 5) show it is necessary to replace approximately 21 to 28 per cent of the milking herd annually. Although there is a relationship between replacement rates and expectation of life in dairy cows, these replacement rates yield no accurate information on the expected life span of cows of different ages.

Lush and Lacy (1) and A. D. B. Smith (4) have each made life expectancy tables for dairy cattle. Lush and Lacy's table represents the condition found in registered herds as their table was estimated from data taken from herd books of each of the five major dairy breeds used in the United States, while Smith's table is based on a study of 2600 Scottish cows. We undertook our study to supplement and enlarge what is already known and also to construct a table that would be representative of dairy animals in Iowa and the midwest, when kept under cow-testing-association conditions.

DATA USED

It is customary in Iowa cow-testing associations to keep records on all cows in the herd with the exception of (a) permanent nurse cows,* (b) valuable purebred cows kept for breeding purposes and not milking at any time during the testing association year, and (c) heifers freshening too young and dried up at once or heifers aborting and dried up immediately.

Therefore, the available records included every cow that had entered the milking herd in any year, regardless of the length of time it remained in the herd.

These records were available for the year 1927–28, for the years 1930–35 and for 22 of the 52 associations operating in the year 1935–36. They

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* A permanent nurse cow is one that nurses a calf throughout the entire lactation period. Cows milked at any time during the lactation period are not considered as permanent nurse cows and are included in the records.

TABLE 1

Age distribution of all cows in Iowa cow-testing associations for years 1927–28 and 1930–36 regardless of the length of time they remained in the herd

Year	Age in years														No record	Total
	2–3†	3–4	4–5	5–6	6–7	7–8	8–9	9–10	10–11	11–12	12–13	13–14	14–15	15 and over		
1927–28	2226	1949	1372	1134	1019	875	742	395	312	179	130	71	36	35	1027	11502
1930–31	8469	5906	4829	4113	3317	2568	1929	1214	900	459	390	150	98	90	4725	39157
1931–32	6514	4446	3529	3047	2655	1844	1418	927	644	382	248	139	84	72	3335	29284
1932–33	4411	3259	2511	1957	1711	1296	966	713	436	247	204	76	69	57	2318	20231
1933–34	4298	3089	2572	1925	1570	1220	1014	608	444	245	163	94	43	39	3177	20501
1934–35	3877	2962	2394	1888	1580	1140	910	649	416	238	175	69	44	32	2286	18660
1935–36*	1652	1124	1051	776	683	528	382	270	186	79	52	36	21	13	1408	8261
Total	31447	22735	18258	14840	12535	9471	7361	4776	3338	1829	1362	635	395	338	18276	147596

* Cows from only 22 of the 52 associations in operation during 1935–36 are included, as the records from the rest of the associations were not available.

† The figures in this column have been corrected by using 82.92 per cent of the original count in order to eliminate an error caused through cows having been recorded twice as two-year-olds on consecutive years. A sample test covering 6538 two-year-old animals showed that 17.08 per cent of them were recorded twice.

covered both purebred and grade cows; the number of purebreds varied upwards from approximately 20 per cent of the total for 1927–28 to 34 per cent for the most recent year.

All cows of each herd were sorted from these records on the basis of age. The summary of this sorting is shown in table 1 which gives the total number of animals of each age for each testing-association year. One column also records the number of cows whose ages were unknown.

Having the number of cows of each age, it is assumed that the difference in numbers represents the mortality of cows as they progress from one age to the next one higher. By mortality of cows, we do not mean that all the cows actually died, but we mean that the cows were removed from the milking herds for one cause or another, such as culling for low production, disease, old age, etc.

By applying the formula $e_x = \frac{T_x}{l_x}$, given by Pearl (3) to the figures representing the total number of cows for each age, the expectation of life for this group of cows was calculated. This formula was used because the cows in each age group were distributed so that it contained cows barely old enough to get into it as well as cows just too young to enter the age group next older. For purposes of comparison, we also calculated the expectation of life for the cows that had passed through the Iowa State College herd during its existence. Table 2 shows the expectations of life for both the

TABLE 2

Expectations of life of cows of different ages in Iowa cow-testing associations and in the Iowa State College dairy herd

Age in years	Expectations of life in years	
	C.T.A. cows	College cows
2– 3	4.1	4.2
3– 4	4.3	4.0
4– 5	4.1	3.8
5– 6	3.8	3.7
6– 7	3.4	3.3
7– 8	3.1	3.1
8– 9	2.7	2.9
9–10	2.7	2.4
10–11	2.4	2.4
11–12	2.5	2.2
12–13	2.0	2.2

cow-testing-association cows and the college cows. The data from the cow-testing associations grouped all cows 15 years and older together. Because some of these cows may have lived to be 18, 19 or more years of age, a discrepancy entered our calculations which was rather large for the older ages, but tended to rapidly lose its force with the younger ages. For that reason, we have extended this table only to cows with ages of 12–13 years.

With one or two exceptions, the life expectancies for the two groups are similar. Perhaps the greatest differences lie in the expectancies for 2–3- and 3–4-year-olds, where the expectancy for the 2–3-year-olds in the cow-testing-association group was less than for the 3–4-year-olds. The extent of the differences is more clearly seen in table 3 which shows the number of

TABLE 3

Number of cows that would be left in the herd at each age for each original hundred 2–3-year-olds providing the animals left the herd in the ratio shown by the age distribution of each group

Age in years	Number of animals at each age	
	C.T.A. herds	College herds
2– 3	100.0	100.0
3– 4	72.3	77.1
4– 5	58.1	57.4
5– 6	47.2	41.7
6– 7	39.9	31.7
7– 8	30.1	22.5
8– 9	23.4	14.8
9–10	15.2	11.2
10–11	10.6	5.2
11–12	5.8	2.8
12–13	4.3	1.2
13–14	2.0	0.8
14–15	1.3	
15 & over	1.1	

cows of each group that would be left in the herd at each age for each hundred 2–3-year-olds which entered the herd, providing animals left the herd in the ratios shown by the age distributions of the two groups.

There were some differences in selecting and sorting the cows in these two groups. Only cows that had freshened were used in the cow-testing-association figures and whenever a cow first freshened, she was called a 2–3-year-old, even though she might not have reached quite the age of two years.

After the counts were completed, it was discovered that some of the animals, probably yearlings and the two-year-olds that calved late in the cow-testing-association year, were entered on the reports as two-year-olds in two consecutive years which increased the counted 2–3-year-olds by that amount. To eliminate this error, a random sample of 6538 two-year-olds was taken from the source data and checked. It was determined that 17.08 per cent of this number had been counted twice on consecutive years. Assuming that this percentage of recounts held true for all the 2–3-year-olds, the original count was reduced by this amount and the corrected figure used in all our calculations.

In sorting the college cows, all two-year-olds were counted whether they had freshened or not, while no yearlings were included, even though they had freshened.

The differences that existed in the selecting and sorting of the cows in the two groups do not account entirely for the 2–3-year-olds of the cow-testing-association group having a shorter life expectancy than the 3–4-year-olds of this same group, while the reverse is true of the college cows. A larger proportion of 2–3-year-olds left the cow-testing-association herds than was the case with the college herd. In the college herd, the recorded reasons for disposal indicate that with few exceptions cows left this herd largely because of disease, bad udders such as the loss of a quarter, and sterility. As would be anticipated under these circumstances, the expectation of life for the 2–3-year-olds is greater than for 3–4-year-olds. If the college herd expectation of life relationship for 2–3- and 3–4-year-olds is the correct one, where little culling other than "forced" culling takes place, then it is apparent that other kinds of culling, such as for low production, have taken place in the cow-testing-association herds. The amount of this culling can be estimated from table 3 for in the college herd, 23 out of each 100 2–3-year-olds were eliminated, while in the cow-testing-association herds, 28 out of each 100 2–3-year-olds left the herds or 5 more than were culled from the college herd. This would indicate that at least 5 per cent of the 2–3-year-olds of the cow-testing-association herds were voluntarily culled by owners.

Some of the years, particularly 1932–35, were affected by the business depression and also drought which caused a shortage of feed. These conditions might have altered the rate and age of culling, but from table 4 it can

TABLE 4

Percentages of 2–3-year-old cows that remained in the cow-testing-association herds to enter each successive year

Year	Age of cows in years						
	2–3	3–4	4–5	5–6	6–7	7–8	8–9
1927–28	100.0	87.6	61.6	50.9	45.8	39.3	33.3
1930–31	100.0	69.7	57.0	48.6	39.2	30.3	22.8
1931–32	100.0	68.3	54.2	46.8	40.8	28.3	21.8
1932–33	100.0	73.9	56.9	44.4	38.8	29.4	21.9
1933–34	100.0	71.9	59.8	44.8	36.5	28.4	23.6
1934–35	100.0	76.4	61.7	48.7	40.8	29.4	23.5
1935–36	100.0	68.0	63.6	47.0	41.3	32.0	23.1
All cows	100.0	72.3	58.1	47.2	39.9	30.1	23.4
	9–10	10–11	11–12	12–13	13–14	14–15	15 and over
1927–28	17.7	14.0	8.0	5.8	3.2	1.6	1.6
1930–31	14.3	10.6	5.4	4.6	1.8	1.2	1.1
1931–32	14.2	9.9	5.9	4.4	2.1	1.3	1.1
1932–33	16.2	9.9	5.6	4.6	1.7	1.6	1.3
1933–34	14.1	10.3	5.7	3.8	2.2	1.0	0.9
1934–35	16.7	10.7	6.1	4.5	1.8	1.1	0.8
1935–36	16.3	11.3	4.8	3.1	2.2	1.3	0.8
All cows	15.2	10.6	5.8	4.3	2.0	1.3	1.1

be seen that except for one year (1927–28) the rate of culling was somewhat the same in all years.

The formula used to calculate the expectations of life applies to populations that are static. It is assumed that replacements within these herds were made through the addition of animals 2–3 years of age or younger. It is recognized that this assumption may not be true as a majority of the herds may have either been increasing or decreasing during the period of the study. Probably some were increasing in size while others were decreasing, so that one would cancel out the other, thus tending to maintain the total number relatively static.

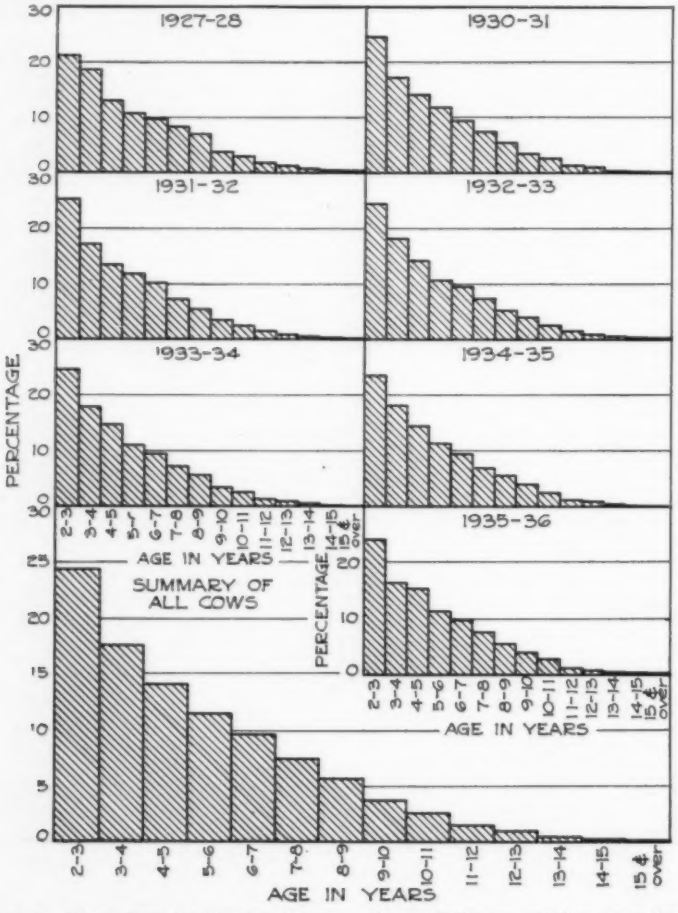

FIG. 1. Percentage distribution by ages of cow-testing-association cows for each year studied and for all cows.

It could happen that instead of the replacements being brought into the herds at 2–3 years of age or younger, that some were purchased at an older age. If this happened, the calculated expectations of life would be changed more as the numbers of such cows increased. Since most of Iowa's dairymen are dairying in order to market the crops from their farms, the replacements for their herds are usually raised on their own farms, tending to keep movement of older cows into herds at a minimum. When purchases are made, they may come from other members of cow-testing associations, in which case there would be no influence on the results.

The expectations of life which were obtained in this study were shorter than those obtained by Lush and Lacy (1) but this can be accounted for partially through the differences in the populations studied. In both the cow-testing-association population and that of the Iowa State College herd, all animals which entered the milking herd were counted, regardless of the length of time they remained in the herd and whether they later calved or not. On the other hand, the populations studied by Lush and Lacy (1) were selected only because they had produced a living calf which was recorded in one of the major dairy breed's herd books. Their data show a tendency for the dams of bulls to be older (2–6 months) than the dams of females. If, in addition, there was a tendency for breeders not to register females from young dams, it would create a bias in the "life expectancies," probably lengthening them.

Frequently, one wishes to know the percentage of cows of each age that make up an average herd. Table 5 gives the percentage distribution by ages of all cows in the cow-testing-association herds used in this study and figure 1 shows not only the percentage distribution by ages of all the cow-testing-association cows for all years but also their distribution for each year.

The table shows that about 56 per cent of all animals in the cow-testing-association herds were under 5 years of age. A rough calculation of the

TABLE 5

Showing percentage of all cows in cow-testing-association herds that are in each age group

Age in years	Percentage
2– 3	24.31
3– 4	17.58
4– 5	14.11
5– 6	11.47
6– 7	9.69
7– 8	7.32
8– 9	5.69
9–10	3.69
10–11	2.58
11–12	1.41
12–13	1.05
13–14	0.49
14–15	0.31
15 or over	0.26

average age of all cows in this group showed the age to be about 4.7 years. This figure is probably slightly low, as in making this calculation, all cows in the group "15 years or over" were given the arbitrary age of 15 years, even though some of the cows were older than this. For comparison, the average age of cows in the college herd was calculated and found to be 4.2 years.

SUMMARY

Iowa cow-testing-association records for seven years yielded information about the ages of 147,596 cows, from which was constructed a table of expectation of life. These expectancies of life were compared with others which were based on the age histories of cows in the Iowa State College herd. Variations existed in these two tables in the life expectancies of 2–3-year-olds and 3–4-year-olds. The 2–3-year-olds of the cow-testing-association group had shorter and the 3–4-year-olds longer life expectancies than these same age groups in the college herd. At other ages, the expectations of life were similar in their relationships.

The low expectation of life for the 2–3-year-olds of the cow-testing-association group was probably due to more culling in these herds at this age than took place in the college herd.

A rough calculation showed the average ages of the cows in cow-testing-association herds to be approximately 4.7 years, and those in the Iowa State College herd to be 4.2 years.

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Student Affiliates:

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Bigott, Gus, Hotel State College, State College.
Brown, Irvan A., Lloydell.
Brunner, J. Robert, 137 Seventh Ave., Royers Ford.
Bryan, Paul, Star Route, Perkasie.
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Hanna, Roy M., Jr., 1905 11th St., Altoona.
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Merry, Walter C., 450 30th St., McKeesport.
Miller, Graham M., 604 W. Union Blvd., Bethlehem.
Miller, Kenneth, 712 Eighth Ave., Bethlehem.

Mowery, Albert S., 115 S. Broad St., Mechanicsburg.
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Snee, E. L., 3735 Brownsville Rd., Pittsburgh.
Sorensen, Howard C., Penn. State Coll., State College.
Turner, Walton L., 54 Amherst Ave., West View, Pittsburgh.
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Crews, Frank, 105 South F. St., Aberdeen.
Drake, John B., Chelan Co. Health Dept., Wenatchee.
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Fuller, Stewart A., Box 23, College Station, Pullman.
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Giberson, Ebert, Dept. Dairy Tech., Montesano.
Golding, N. S., State Coll. of Wash., Pullman.
Goodwin, Dwight Alden, Box 6134, Hillyard Sta., Spokane.
Herrington, Fred L., 402 E. 29th St., Vancouver.
Hill, Otto J., Extension Service, Pullman.
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Krassin, C. W., Box 482, Burlington.
Kristensen, A., 1440 Jackson St., Seattle.
Larson, Harold, 21 W. Lee, Apt. 104, Seattle.
Miller, Ray, 503 Smith Tower, Seattle.
Nelson, Hans, Jr., Pine Manor, 202 Washington St., Pullman.
Olsen, John E., Health Dept., Court House, Shelton.
Palmer, W. Horace, Box 250, Steilacoom.
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Harris, Robert J., 1112 Racine St., Janesville.
Harris, Roy T., Dairy Records Office, Madison.
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Ireland, Wm. R., 1222 Munroe Ave., Racine.
Jackson, H. C., Univ. of Wis., Madison.
Kaeser, W. G., 110 Ely Place, Madison.
Kelly, Frank W., 620 N. 8th St., Milwaukee.
Knoll, Roy P., 1633 N. 13th St., Milwaukee.
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Schmit, Ray, 2206 N. 30th St., Milwaukee.
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Rive, Henry, Dept. of Agric., Victoria, B. C.
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Singleton, John Franklin, Confederation Bldg., Ottawa, Ont.
Skinner, H. C., 122 Richmond St. West, Toronto, Ont.
Smallfield, H. A., Ontario Agric. College, Guelph, Ont.
Sproule, W. H., Ontario Agric. College, Guelph, Ont.
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Board of Health Laboratories, 763 Hornby St., Vancouver, B. C.
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ERRATA

April, 1939, page 218, reference (1) should read: Barthel, Chr., Weitere Untersuchungen über die Reduktaseprobe, sowie Vergleiche mit einigen anderen neueren milchhygienischen Untersuchungsmethoden. Zschr. Untersuch. Nahrungsmitt. 34: 137–164. 1917.

April, 1939, page 248, reference (16) should read: Fürth, O. von, and Charnass, D., Über die quantitative Bestimmung der Milchsäure durch Ermittlung der daraus abspaltbaren Aldehydmenge. Biochem. Zschr. 26: 199. 1910.

April, 1939, page 249, reference (33) should read: Scheunert, A., and Pelchrzim, H. von, Über den Gehalt des Blutes verschiedener Tierarten an Zucker, Rest-N, Harnstoff-N, Kreatininkörpern und Harnsäure nach den Folinschen Methoden. Biochem. Zschr. 139: 17–29. 1923.

April, 1939, page 249, reference (45) should read: Abderhalden, E., Zur quantitativen vergleichenden Analyse des Blutes. Z. Physiol. Chem. 25: 65. 1898.

June, 1939, page 436, line 25 should read: The proteins of corn gluten meal do not appear to entirely make good the cystine deficiencies of alfalfa crude protein.

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ABSTRACTS OF LITERATURE

BACTERIOLOGY

531. A Microbiological Assay for Riboflavin. E. E. Snell and F. M. Strong, Univ. of Wis., Madison, Wis. Ind. Eng. Chem., Analyt. Ed., *11:* 6, 346–50. 1939.

A biological method is described for estimating the riboflavin in natural materials by determining the amount of growth of *Lactobacillus casei* in a medium containing the unknown sample. To a riboflavin-free basal medium is added a prepared extract of the unknown, the mixture is inoculated and incubated and the amount of growth determined by measurement of acidity or turbidity of the culture. The reliability of the method is supported by agreement of the assay results at different levels, recovery of added riboflavin, successful determination of riboflavin in the presence of photolyzed extracts, and specificity of structure required for activity. A series of assays on the same product at two-week intervals yielded results which compared well.

B.H.W.

532. Influence of Bacteria on Oxidation of Ascorbic Acid. William B. Esselen, Jr., Dept. of Bacteriology and Physiology, Mass. State College, Amherst, Mass. Food Research, *4:* 4, 329. 1939.

As a result of studies with forty-five different strains of bacteria (39 species), it was concluded that most of the organisms significantly retarded oxidation of ascorbic acid, especially when they were cultured in media containing readily fermentable carbohydrates. No evidence was found that bacteria are destructive to ascorbic acid, nor did any of the organisms studied appear to synthesize the vitamin.

F.J.D.

BUTTER

533. Factors Affecting Amount of Mold Mycelia in Butter. J. Adams and E. H. Parfitt, Purdue Univ., Lafayette, Ind. Nat'l Butter and Cheese J., *30:* 10, 29. 1939.

When initial contamination is held constant, mold mycelia fragments in butter are an index of the length of time, temperature of storage, and the oxygen relationship of the cream from which the butter was made. Manufacturing operations do not influence the mold mycelia content of butter.

W.V.P.

534. Composition Control with Special Reference to the Rolless Churn. C. C. Totman, S. Dakota State College, Brookings, S. D. Nat. Butter and Cheese J., *30:* 8, 12. 1939.

The roll-less churn produced butter of remarkably uniform composition.

W.V.P.

CHEESE

535. A Study of Winter and Summer Made Cheddar Cheese. J. C. MARQUARDT, N. Y. Agric. Exp. Sta., Geneva, N. Y. Nat. Butter and Cheese J., *30:* 9, 28. 1939.

Cheese made from winter milk is "low quality" because it lacks flavor when cured at low temperatures. Ten lots of winter-made raw-milk cheese and 9 lots of summer cheese from New York state factories were scored, analyzed and given a cheese flavor index value during ripening; four lots of winter-made Canadian cheese and 7 lots of Wisconsin summer cheese were given the same inspection. All were cured at 35 to 40° F. Like studies were conducted using cheese made experimentally from high-quality, raw and pasteurized milk. Some of the experimental cheeses were cured at 35 to 40° F., others at 55° F. after holding 2 weeks at 35 to 40° F., the remainder were cured at 55° F. The factory cheese made in summer developed greater flavor intensity in a shorter time than did the cheese made in winter; the quality of the two types as indicated by the average scores was quite similar. The experimental cheese made in summer showed the same seasonal effects. Curing at low temperatures delayed good as well as bad flavor development but produced cheese of better quality. Curing raw-milk cheese first at high then low temperature lowers the flavor and body score and produces off flavor development which makes it difficult to evaluate flavor intensity. Combinations of low and high temperature curing are suggested to force cure. W.V.P.

CHEMISTRY

536. The Concentration of Ionized Magnesium and Calcium in Milk. R. NORDBÖ, Dept. of Physiology, Univ. of Oslo, Oslo, Norway. J. Biol. Chem., *128:* 745. 1939.

The results of analysis of Na, K, Ca, Mg, Cl, inorganic phosphate, and citric acid and of the determination of H and Mg indicate a value of the ionic strength of about 0.075 to 0.08 in ultra filtrate of fresh market milk. The K of calcium citrate was found to be 3.16×10^{-4} and of magnesium citrate 2.65×10^{-4}, at 37° and ionic strength of 0.08. Lactose combines with calcium and magnesium to form non-ionized compounds. The concentration of magnesium ions in ultra filtrates of fresh market milk is 0.4 to 0.5 mM. per liter. Total diffusible Mg is about 2.5 mM. per liter, of the diffusible Ca of fresh market milk (8.5 to 10 mM. per liter) only about 20 per cent is present as calcium ions. K.G.W.

537. Kniaseff Fat Test. HORACE H. SELBY AND THELMA A. SELBY, Hage's, Ltd., San Diego, Calif. Ind. Eng. Chem., Analyt. Ed., *11:* 7, 393–94. 1939.

The fat test of Kniaseff was found to compare with the Mojonnier fat test on ice cream and ice milks more favorable than any of 25 other modifications of the Babcock method which were investigated by the authors. The method is essentially one of alkali-alcohol-ether extraction carried out in the conventional Babcock cream test bottle by the addition to the sample of two reagents made up according to the directions given. The loaded bottles are centrifuged, the fat column read and a correction applied which gives results generally within 0.1 per cent and always within 0.4 per cent of values obtained by the Mojonnier method. B.H.W.

538. Xanthine Oxidase: Purification and Properties. ERIC G. BALL, Kaiser Wilhelm-Institut für Zellphysiologie, Berlin-Dahlem, Germany. J. Biol. Chem., *128:* 51. 1939.

A process is described whereby an enzyme preparation is obtained from whole milk, and which is 500 times more active per unit of dry weight than the original material. Evidence is presented that this enzyme is a flavoprotein. K.G.W.

539. Determination of Riboflavin in Milk by Photoelectric Fluorescence Measurements. DAVID B. HAND, Cornell Univ., Ithaca, N. Y. Ind. Eng. Chem., Analyt. Ed., *11:* 6, 306–09. 1939.

A fluorometric method for determining the riboflavin in milk or whey which is rapid, accurate, and specific is described. Fifty ml. of acetone is added to 25 ml. of milk and the fluorescence of the filtrate is compared with that of a cube of uranium glass. The fluorescence intensities of samples of riboflavin were determined and the uranium glass was calibrated against solutions containing known amounts of the pigment. The apparatus developed for making the fluorescence measurements consisted of a photoelectric cell, a microameter, a capillary mercury lamp, two glass filters, a block of uranium glass and a square glass bottle. B.H.W.

DISEASE

540. Relationship between Carotene, Blindness Due to Constriction of the Optic Nerve, Papillary Edema and Nyctalopia in Calves. L. A. MOORE. J. Nutrit., *17:* 443–459. 1939.

For the past 20 years various investigators have noted a peculiar type of blindness in calves associated with rations where poor quality or no roughage was fed. Previous work has shown that the blindness is due to a constriction of the optic nerve where it passes through the optic foramen caused by a stenosis of the bony canal. Further work with calves has shown that the blindness is preceded by nyctalopia, and ophthalmoscopic observations revealed papillary edema and bleaching of the Tapetum Lucidum. Hol-

stein calves placed on the blindness producing ration and fed crystalline carotene dissolved in cottonseed oil in quantities sufficient to keep the blood plasma carotene level well above a lower limit of 0.13 micrograms per ml. did not develop these eye changes. Vitamin A deficiency is therefore the causative factor.

In mature cows nyctalopia and papillary edema develop but blindness due to constriction of the optic nerve does not develop due to the fact that the bony optic canal has already been formed. In calves blindness due to constriction of the optic nerve and papillary edema are associated, while nyctalopia is a separate process but due to the same deficiency.

Papillary edema is a cardinal sign of increased intracranial pressure. Vitamin A may therefore be necessary for the regulation of this pressure. Increased intracranial pressure may also account for certain nervous manifestations of vitamin A deficiency. C.F.H.

FEEDS AND FEEDING

541. Carotene Content of Different Varieties of Green and Mature Soybeans and Cow Peas. W. S. Sherman and W. D. Salmon, Labratory of Animal Nutrition, Ala. Polytechnic Inst., Auburn, Ala. Food Research, *4:* 4, 371. 1939.

Most varieties of soybeans in the green stage where found to be very good sources of carotene ranging from 212 to 705 γ per 100 grams of fresh sample. The varieties of cow peas studied contained from 140 to 231 γ per 100 grams of fresh sample. The carotene content of mature soybeans ranged from 8 to 243 γ per 100 grams of air-dry sample. All varieties of mature cow peas were low in carotene, being comparable to the lowest soybean varieties. Reduction of as much as 50 per cent in carotene resulted from allowing plants to stand in the field two or three weeks after the seeds had reached maturity. F.J.D.

542. Beobachtungen über den Einfluss verschieden grosser Heugaben auf Kraftfutterverbrauch und Gedeihen der Kälber. (Observations on the Influence of Varying Amounts of Hay on Consumption of Concentrates and Growth of Calves.) L. Paloheimo, Institut für Haustierlehre der Universität Helsinki. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *9:* 4–6, 287–293. 1937.

A continuation of work reported in earlier numbers of this Journal (9, 52 and 234).

When skim milk (plus cod liver oil) was fed alone, the amount given at individual feedings was of significance in the growth of the calves, for when the portions were small the ill effect of lack of roughage became apparent in the second month, whereas when large individual portions of skim milk were fed this effect was not evident until the fourth month.

If no hay was fed, larger quantities of ground oats were eaten, but the quantity of corn meal eaten was unchanged. The calves suffered as much when fed skim milk and corn meal as when fed skim milk alone.

When no hay was fed and ground oats was the concentrate the calves throve nearly as well as when both hay and ground oats were fed. Ground oats seems to be a natural mixture of roughage and concentrate.

With limited hay feeding the calves' requirements for concentrates were the same with either corn or oats. If hay was offered in abundance the consumption of ground oats was limited more sharply than that of corn meal. If all constituents of the ration were offered *ad lib*, consumption of hay was about the same regardless of whether the concentrate was corn meal or ground oats. J.G.A.

543. Die wissenschaftliche Grundlage des stärkwertes. (The Scientific Basis of Starch Values.) J. AXELSSON, Institut für Haustierfütterung der Landw. Hochschule Schwedens, Uppsala. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *11:* 3, 176–205. (2 Abbildungen.)

A review and restatement of the problem in the light of recent additions to knowledge of the subject. Optimal contents of crude fiber in the dry matter and of protein in the digestible organic substance are necessary if metabolizable energy available for gains in live weight is to be utilized as completely as possible. Eighteen to 23 per cent of crude fiber and 10 to 18 per cent of protein are considered optimal. Therefore, when the effect of a ration is to be determined by difference, both the basal ration and the feed (or ration) on trial must contain optimal quantities of fiber and protein. As this was not the case in Kellner's work his starch values are open to serious criticism. Information concerning the fiber and protein content of rations regardless of the feeds which compose the rations is, however, not sufficient. Care must be taken that the content of other substances with special nutritive values is optimal. Because of this, relative values of feedstuffs cannot be determined on a net calorie basis but instead must be judged by the energy content at the beginning of the production process. Content of metabolizable energy seems to deserve preference over content of digestible energy. J.G.A.

544. Die stimulierende Wirkung des Proteins im Tierfutter. (The Stimulating Effect of Protein in Feeds.) J. AXELSSON, Institut für Haustierfütterung der Landw. Hochschule Schwedens, Uppsala. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *11:* 3, 162–175. (3 Abbildungen.)

A review of the more recent work on the subject. The author summaries as follows:

Gains in weight increase and simultaneously feed energy is better utilized when the protein content of the feed is raised to a certain degree (not stated). A further increase in protein causes a decrease in gains and also in energy utilization. The optimal protein content for gains in weight diminishes with the growth of the animal. Utilization of the protein for deposit begins to decrease long before the optimum for gains in weight is reached, and continues to decrease after this point is surpassed. Simultaneously a definite and continuously increasing part of the nitrogen and energy of the feed is being excreted in the urine. Thus the favorable effect of the protein, when offered in optimal quantity, is manifest, even though only a reduced amount of it is stored in the body. Therefore a stimulating effect must be assumed.

During lactation also there seems to be an increased utilization of feed energy when protein is increased, and that beyond the quantity of protein needed for nitrogen balance in the body. The optimal quantity seems to be higher than hitherto has been supposed. Whether the protein content necessary for highest milk yield corresponds to that for best energy utilization is still in doubt. As a result of this change in viewpoint on effect of protein, the deciding factor in formulating protein standards for practical use must be the utilization of the total ration rather than that of the portion used only for production. J.G.A.

545. Über einen Einsäuerungsversuch mit Zusatz von Ammonbicarbonat und Fütterungs versuch mit dem erhaltenen Gärfutter an Milchkühen. (Experiments on the Making and Feeding to Milch Cows of Ensilage with Added Ammonium Bicarbonate.) C. WINDHEUSER, Landesversuchsanstalt für landw. Chemie, Landw. Hochschule Hohenheim, Biedermann's Zentralblatt, Abteilung B., Tierernährung, *11:* 2, 139–146. 1939.

German yellow maize was fermented in three silos, NH_4HCO_3 being added as small crystals to the contents of one silo at chopping time. All the silages were of good quality, and were fed to milking cows to ascertain whether the nitrogen of the ammonium salts formed in the process (chiefly lactate and acetate) could replace a portion of the food protein.

Effect on milk yield was noted of substituting 51.3 grams of N from ammonia in 15 kilos of silage for 47.8 grams of N from the digestible protein in 1.2 kilo of rape cake. No effect of the nitrogen from ammonia could be noticed; in one period when silage without ammonium salts was fed, and rape cake was omitted, milk yield was not significantly lower than when silage with ammonium salts was fed. J.G.A.

546. Zur Beifütterung von gedämpften Kartoffeln an Saugkälber. (Feeding Steamed Potatoes to Young Calves.) H. STEINHARDT, Agrikulturchemischen und Bakteriologischen Institut der Schlei-

sischen Friederich-Wilhelms-Universität. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *11:* 2, 66–120. 1939. (3 Abbildungen.)

Ten calves fed whole milk, steamed potatoes and malt were compared with six calves fed whole milk alone. Metabolism balance trails beginning after one week on the ration and continuing for four weeks were conducted with seven of the group receiving potatoes and with four of the controls.

Digestive troubles were experienced with four of the ten calves fed potatoes; two of them recovered. Cases of diarrhea were not considered to be caused by the potatoes.

Data are given for digestibility and starch values of both rations. The potatoes had a distinct protein sparing effect, and the animals made greater gains in live weight than those on milk alone. Detailed directions are given for preparing and feeding the potato-malt-milk gruel. J.G.A.

547. Untersuchungen über den Wert von Harnstoff, Glykokoll, Amidflocken und-schnitzeln für Milchkuhe und wachsende Schafe. (Studies of the Value of Urea, Glycocoll, Amide Flakes (Potato) and Amide Cuttings (beet) for Milch Cows and Growing Sheep.) W. KIRSCH UND F. SAUER, Tierzucht-Institut der Universität Konigsberg. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *10:* 6, 451–480. 1938. (1 Abbildung.)

Several digestion trials were conducted with cows and one with sheep; some of the trials were prolonged and nitrogen balances were ascertained in every case.

If urea and glycocoll are fed in the pure form they must be introduced into the ration very gradually and carefully, otherwise health disturbances and marked reduction of milk yield will result.

When urea was fed in combination with potato flakes or beet cuttings, the mixture was readily eaten in quantities of 1.2–1.5 kilos and proved entirely beneficial.

Urea nitrogen was completely assimilated by both cows and sheep regardless of whether the urea was fed as such or in combination with the amide flakes or cuttings.

In the trials with cows, when the urea-amide-flake or cutting mixture was substituted for ground soybeans so that the urea nitrogen amounted to 20 per cent of the total nitrogen intake, the urea nitrogen was found to have a value for productive metabolism equivalent to from 47–66 per cent of the value of the nitrogen in the basal ration. J.G.A.

548. Über die Einwirkung der Eiweissunternährung auf die Milchproduktion frischmelkender Kühe. (The Effect of Limited Protein Feeding on the Milk Production of Fresh Cows.) P. KAJANOJA,

Landwirtschaftliche Institut Mustiala in Finnland. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *9:* 4–6, 525–530. 1937.

Cows that had recently calved and were fed considerably less protein than is called for by generally accepted feeding standards, in most cases showed no decrease in milk yield as compared with their records in former years. Nor did an increase in the protein of the ration have any effect in raising their production. Only when the amount of protein supplied was so small that the cows received only as much or about as much true protein as they were secreting in their milk, was a decrease in milk production noted. In such cases an increase in the protein was followed by an increase in milk yield.

These facts indicate that the original ability of wild cattle to take protein from their body substance (chiefly from their muscles) to sustain milk production, still exists in the highly bred races of present day dairy cows to an extent scarcely to be expected.

A very small supply of protein seemed to effect a decrease in fat content of the milk, and conversely, increasing the protein in the ration resulted in increased fat production. Thus it is seen that fat content is influenced more than quantity of milk by a reduction in the protein allowance.

J.G.A.

549. Über die Ensilierung unter Zusatz von Molkereiabfall-produkten und über die Korrelation zeischen pH, Ammoniakzahl, Buttersauregehalt und Geruch bei Grassilagen. (Ensiling with Additions of Dairy By-Products and Correlation between pH, Ammonia Number, Butyric Acid Content and Odor in Grass Silages.) E. Brouwer, Physiologischen Abteilung der Rijkslandbouwproefstation in Hoorn, Niederlande. Biedermann's Zentralblatt, Abteilung B., Tierernährung *9:* 4–6, 508–524. 1937. (9 Abbildungen.)

One hundred twenty-one samples of silage (86 of them from grass) which had been ensiled with whey and other dairy by-products, were examined.

Most of the samples were of inferior quality. Evidently protein-rich crops and Netherland young grass are too high in protein and too low in sugar to permit satisfactory preservation by addition of whey, etc.

Frequency curves of the pH values, the butyric acid contents, and the ammonia fractions (ammonia N as per cent of total N), appeared to be polymodal, even if only one crop (grass) was considered. The polymodality is the expression of different bacteriological processes.

On the average the pH values decreased as the samples contained less protein in the dry matter and less moisture. One per cent of moisture more or less corresponded in grass silage with 0.03 pH units; one per cent

of crude protein (ammonia included as $6.25 \times N$) in the dry matter more or less with 0.07 pH units.

The ammonia content increased considerably with rising pH. At pH 4.0 in grass silage an average of 8 per cent of the total nitrogen was present as NH_3, at pH 5.0 it reached 24 per cent and at higher pH values correspondingly higher NH_3 values were observed.

Butyric acid in the grass silage was very low at pH 4.0; above this value the pH and the butyric acid rose simultaneously and by about the same number of units.

In grass silage especially the odor gives a rather good idea of the pH range and the bacteriological type. In judging the quality of silage for practical purposes, in many instances a simple examination of the odor and the pH is sufficient, although it is not to be implied from this that more detailed investigation is valueless. J.G.A.

550. Der Wert der Amide bei der Fütterung des Rindes. Historische Betrachtung der Entwicklung der Amidfrage, kritische Wertung des Standes unserer heutigen Kenntnisse. (The Value of Amides in the Feeding of Cattle. An Historical Review of the Development of the Amide Problem, and a Critical Evaluation of the Status of our Present Knowledge.) K. KREBS, Institut für Tierzuchtung und Haustiergenetik der Universität Berlin. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *9:* 4–6, 394–507. 1937.

A comprehensive review with a bibliography of 126 references. From a summary of all the evidence the author concludes that the system of rationing farm animals based on digestible crude protein is unsound, since the function of the non-protein nitrogen fraction is not comparable to that of the protein moiety. Although non-protein nitrogen must be taken into account, it must be clearly distinguished from protein nitrogen. J.G.A.

551. Über die Bestimmung des verdaulichen Rohproteins auf Künstlichen Wege. (The Determination of Digestible Crude Protein by Artificial Means.) P. SCHWARZE, Kaiser-Wilhelm-Institut für Züchtungsforschung Müncheberg/Mark. Biedermann's Zentralblatt, Abteilung B., Tierernährung, *9:* 4–6, 346–354. 1937.

A report of trials to ascertain the digestibility of crude protein in fodder plants, especially legumes. Digestion was effected with pepsin-hydrochloric acid and pancreatin-soda. Trichloracetic acid was used to precipitate the indigestible nitrogen compounds. In several cases control experiments were run with rats.

Hydrolysis and digestion in the organism is incomplete; the greater part of the excrement nitrogen is precipitated by trichloracetic acid and is in turn largely hydrolyzed by proteolytic enzymes.

Under the conditions of the experiment it has been possible to classify nitrogen compounds into those which are hydrolyzed by proteolytic enzymes and those which distinctly are not.

In leguminous seeds and rye digestibility was complete or nearly so, while in alfalfa plants, and corn and barley (grain) under the same conditions a residue was left which was not precipitable by trichloracetic acid. The nature of this residue could not be determined; it may or may not be protein. J.G.A.

552. Determination of Carotene in Silage. An Improved Method. D. M. Hegsted, J. W. Porter and W. H. Peterson, Univ. of Wis., Madison, Wis., Ind. Eng. Chem., Analyt. Ed., *11:* 5, 256–58. 1939.

An improvement in the accuracy of the carotene determination of silage whereby pigments other than carotene are removed from the silage extracts by the use of a diacetone solution is reported. The presence of foreign pigments in the carotene solution has heretofore often caused the determinations by the old Willstätter-Stoll method and its subsequent modifications to be too high. The authors have substituted a solution of 100 volumes of diacetone and 6 volumes of water for the usual 90 per cent methyl or 85 per cent ethyl alcohol solution used to remove foreign pigments from the carotene solution. Comparative data are given for the alcohol and acetone methods. The solutions from both methods were subjected to chromatographic analysis. B.H.W.

FOOD VALUE OF DAIRY PRODUCTS

553. The Influence of Different Levels of Milk Intake on the Acid-Base Mineral Balances of Children. H. A. Hunscher, F. C. Hummel and I. G. Macy. J. Nutrit., *17:* 461–471. 1939.

The nitrogen, acid base and excess base balances are given for seven healthy children, ages 5 to 10 years, when they were consuming 400, 600, 800, and 1,000 gm. of milk daily. All the children were storing both acid and base throughout the entire study but greater quantities were stored with larger intakes of milk. C.F.H.

554. The Utilization of the Calcium of Milk by Pre-School Children. G. Kinsman, D. Sheldon, E. Jenson, M. Bernds, J. Outhouse and H. H. Mitchell. J. Nutrit., *17:* 429–441. 1939.

Data were presented from a study of the calcium metabolism of five pre-school girls and six pre-school boys. The calcium level in the diet was increased by increasing the milk solids in the diet. The mean value for the calcium utilization of the eleven subjects was 19 per cent. The limited ability of the pre-school child to utilize calcium is entirely different from results with rats. C.F.H.

555. The Utilization of the Calcium in Various Greens. MARY SPEIRS, J. Nutrit., *17:* 557–564. 1939.

The calcium in turnip greens was about as available as that in milk, while that in tender green collards and kale was slightly less available. Not only was the calcium in New Zealand spinach utilized poorly, if at all, but also the utilization of the calcium of the milk in the diet was diminished by the presence of the New Zealand spinach. C.F.H.

ICE CREAM

556. Whipping Capacity of Ice Cream Mixes. ALAN LEIGHTON AND ABRAHAM LEVITON, Bu. Dairy Ind. U. S. D. A., Washington, D. C. Ind. Eng. Chem., *31:* 779. June, 1939.

The effect of variations in manufacturing procedure and mix ingredients upon the whipping properties of ice cream mix is graphically presented. The whipping capacity of each mix is shown by an overrun line in which the highest overrun obtainable at a given temperature is plotted against that temperature. The resulting straight line is an equilibrium line which is characteristic of every mix of a given composition.

The overrun lines of many mixes are presented. They show that the aging of a mix in which butterfat and improvers are absent will increase its shipping properties if the mix has been heated; that an increase in homogenization pressure increases stability and maxium overrun while an increase in homogenization temperature under some conditions accelerates aging. Butterfat is an overrun depressant, especially when the fat is derived from butter, but if the butter is homogenized with skim milk and the reconstituted cream again homogenized with the mix, whipping properties are greatly improved. There is some tendency of sugar to raise overrun but this is offset by its detrimental effect upon stability. Gelatin and carob bean extract have similar effects on the overrun line. Sodium alginate acts as a dual stabilizer, stabilizing the structure and the fat emulsion.

Studies on the temperature-overrun relationship indicate proper adjustment of the freezing process, will lower freezing time, and clarify existing ideas concerning the whipping properties of mixes. B.H.W.

557. A Comparison of Gelatin and Sodium Alginate as Stabilizers in Ice Cream. P. H. TRACY AND S. L. TUCKEY, Dept. of Dairy Husbandry, Univ. of Ill., Urbana, Ill. Food Research, *4:* 4, 335. 1939.

When sodium alginate and gelatin were used in optimum amounts in ice cream there was no decided preference for either stabilizer as far as the body of the ice cream or the whipping time was concerned. No differences in flavor were detectable and no off-flavors attributable to the stabilizers were encountered. Melt-down trials showed ice cream containing sodium

alginate to melt more rapidly and to have a smoother appearance than that containing gelatin. F.J.D.

MILK

558. Irradiated Milk. Vitamin D Potency as a Function of Energy Input. BRIAN O'BRIEN, H. DOUGLAS MCEWEN AND KENNETH MORGAREIDGE. Ind. Eng. Chem., *30:* 7, 839–842. 1938.

The results of a study on the antirachitic activation of whole milk by direct action of ultra-violet energy from carbon arcs is presented. A theory is developed which gives a means of predicting the amount of energy input to the arc necessary to produce a given vitamin D potency in the treated milk. As the energy input increases the vitamin content of the milk approaches a maximum value. With continued irradiation the peak in antirachitic potency is passed and a gradual decline occurs with slow decomposition of the active principle. B.H.W.

559. Cereal Flours as Antioxidants in Dairy Products. W. S. MUELLER AND M. J. MACK, Dept. of Dairy Industry, Mass. State College, Amherst, Mass. Food Research, *4:* 4, 401. 1939.

Studies revealed that oat flour, whole oat flour, and corn flour have similar antioxidative properties when used in milk. Wheat (blended and whole) barley, rye and rill flours possess little antioxidative value. In ice cream, corn flour gives similar antioxidative protection to that given by oat flour and has more effect than the latter as a stabilizer. F.J.D.

560. The Irradiation of Milk. K. G. WECKEL AND H. C. JACKSON, Univ. of Wis., Madison, Wis. Wis. Agr. Exper. Sta. Res. Bull. 136. March, 1939.

Irradiation of milk is a process of exposing milk under proper conditions to radiant energy to increase its antirachitic value (vitamin D potency). Milk is a good carrier of vitamin D because it contains a desirable balance of the antirachitic minerals, calcium and phosphorous. Milk has vitamin D as a natural constituent which varies with several factors such as individual cows, feed, and exposure of cows to sunlight. Radiant energy from sunlight has wave lengths which can change certain substances to vitamin D. The chief activatable substance in milk is believed to be 7-dehydrocholesterol. Artificial sources of radiant energy to do this work are carbon arc lamps, quartz mercury vapor lamps, and cold quartz lamp tubes. Radiation from these sources differs quantitatively and qualitatively. Types of equipment designed to use these sources are illustrated and described. Methods of controlling the process of irradiating such as milk flow and radiation intensity are discussed. Factors affecting efficiency of irradiation are inter-

related. Those mentioned are milk composition, concentration of activatable substances, opacity of milk; thickness, velocity and turbulence of milk film; intensity and composition of radiation; and mechanical factors affecting characteristics, capacity, and dimensions of the film as well as distance from the source of and angle of incidence of radiation.

Commercial irradiation does not destroy vitamins A, B or G, is only indirectly responsible for slight, variable reduction of C, and reduces slightly the bacterial content. Milk unduly exposed to ultra-violet light may develop a typical "activated" flavor which is associated with the protein fractions of milk. Radiation with other than the antirachitic wave lengths, though not as effective, may produce this flavor. Normal irradiation procedures cause no detectable changes in composition or digestibility of milk. This article is 55 pages in length, contains 54 figures, and 85 references.

W.V.P.

PHYSIOLOGY

561. Influence of the Hypophysis on Carbohydrate Metabolism. SAMUEL SOSKIN, H. LEVINE AND W. LEHMANN, Dept. of Metabolism and Endocrinology, Michael Reece Hospital, and the Dept. of Physiology, Univ. of Chicago. Am. J. Physiol., *127:* 3, 463. 1939.

An anterior pituitary extract (Phyone) which was shown to be capable of maintaining the carbohydrate levels of fasting hypophysectomized animals, was studied for its effect on the utilization of carbohydrate.

This hormone maintains blood sugar level by stimulating the liver to gluconeogenesis rather than by acting upon the extra-hepatic tissues of the body. The reduction of hyperglycemia, glycosuria and ketosis in the depancreatized animal following hypophysectomy, is due to a decrease in the rate of hepatic gluconeogenesis, and not to a greater utilization of the sugar by the extra-hepatic tissues. D.L.E.

562. Significance of Amino Acids for the Maintenance of Nitrogen in the Adult White Rat. PAUL A. WOLF AND RALPH C. CORLEY, Laboratory of Biochemistry, Purdue Univ., Lafayette, Ind. Am. J. Physiol., *127:* 3, 589. 1939.

Previous work has shown that the nitrogen of mixtures of amino acids would spare completely that of the tissues of rats, fed diets low in protein. The present work indicates that adult female white rats also require for maintenance of nitrogen balance each of the amino acids essential for growth. Rations practically devoid of protein but supplemented with vitamin preparations and mixtures of the nine amino acids essential for growth, histidine, isoleucine, leucine, lysine, methionine, phenylalanine, threonine, tryptophane and valine, have proven satisfactory for maintaining nitrogen equilibrium in the growing as well as in the adult animal. D.L.E.

563. Further Investigations Concerning the Effects of Formalin on the Gonadotropic and Thyroid-Stimulating Hormones of Cattle Anterior Pituitary Glands in the Guinea Pig. LEO LOEB AND S. G. HAYWARD, Laboratory of Research Pathology, Oscar Johnson Institute, Washington University School of Medicine, St. Louis, Mo. Am. J. Physiol., *127:* 3, 497. 1939.

Immersion of single cattle pituitary glands in formalin solutions causes a marked reduction or complete suppression of the leuteinizing and atresin effects and in many instances permits a full maturation of follicles in the ovary of the injected. guinea pig. The largest number of positive results were obtained if heifer pituitaries were immersed in 0.25 per cent solutions of formalin at pH 5 for seven days.

It is possible to maintain cattle pituitary glands, which have been converted by the action of formalin into maturation-producing and estrogenic-agents in the sex organs of the guinea pig, unchanged for at least four months, but presumably for even longer periods by immersion in glycerin.

The thyroid hormone is injured or destroyed by formalin. This action is independent of the leuteinizing or atresin effects which are produced in the ovaries. This confirms the conclusion that leuteinizing, atresin and thyroid-stimulating actions are not due to the same agent. D.L.E.

564. The Role of Sex, Estrogenic Hormone, Fasting and Diuresis in the Response of Crop Sacs of Pigeons to Prolactin. ROBERT W. BATES, OSCAR RIDDLE AND ERNEST L. LAHR, Carnegie Institution of Washington, Sta. for Exper. Evolution, Cold Spring Harbor, N. Y. Am. J. Physiol., *127:* 3, 422. 1939.

When prolactin is injected subcutaneously into immature pigeons, the crop-sacs of males usually give a greater (growth) response, but following intramuscular injections of prolactin there is little or no sex difference in response.

The apparent antagonistic action of estrogen against prolactin is found thus to be largely though probably not wholly the result of the diuresis and reduced food intake which dihydroestrone induces in pigeons; any additional or residual effect is thought likewise to rest not upon a specific antagonism but upon one or another of the general effects of dihydroestrone on metabolism.

Estrogen injected simultaneously with prolactin into pigeons inhibits the gains in body weight which, like the response of growth in the crop sacs, characterize the normal action of prolactin in pigeons; and this inhibition of gain in body weight is apparently produced in the same indirect ways as is the inhibition of the crop-sac response. D.L.E.

565. The Role of the Pituitary in the Calorigenic Action of Vitamin D. A. BARTOLI, J. FELDMAN AND C. I. REED, Dept. of Physiology,

Univ. of Ill. College of Medicine. Am. J. Physiol., *127:* 3, 552. 1939.

The calorigenic response of normal dogs to doses of vitamin D do not occur following hypophysectomy. Both the pituitary and the thyroid are necessary for the calorigenic response to vitamin D. The calorigenic response is largely independent of the effect of vitamin D on mineral metabolism. D.L.E.

566. The Partition of the Serum Globulins of the Dairy Goat. E. P. Reineke, V. E. Peterson and C. W. Turner, Dept. of Dairy Husbandry, Univ. of Mo., Columbia, Mo. J. Biol. Chem., *128:* 1. 1939.

A study of the (sodium sulfate) fractionation pattern of the serum globulin complex of the goat revealed the following fractions: euglobulin, pseudoglobulin I, and pseudoglobulin II. Subdivision products of the latter two groups were obtained. K.G.W.

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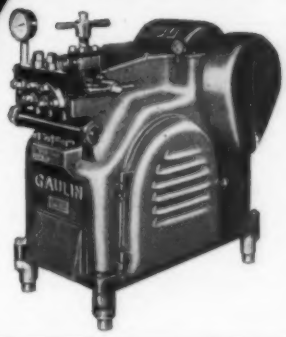

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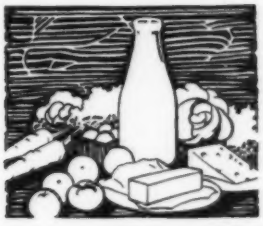

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